Cell Organoid

Review

https://doi.org/10.26599/CO.2026.9410024

Brain organoids and brain disease organoids modeling in the age of artificial intelligence

Authors

Jianfei Long*, Xinxin Han*

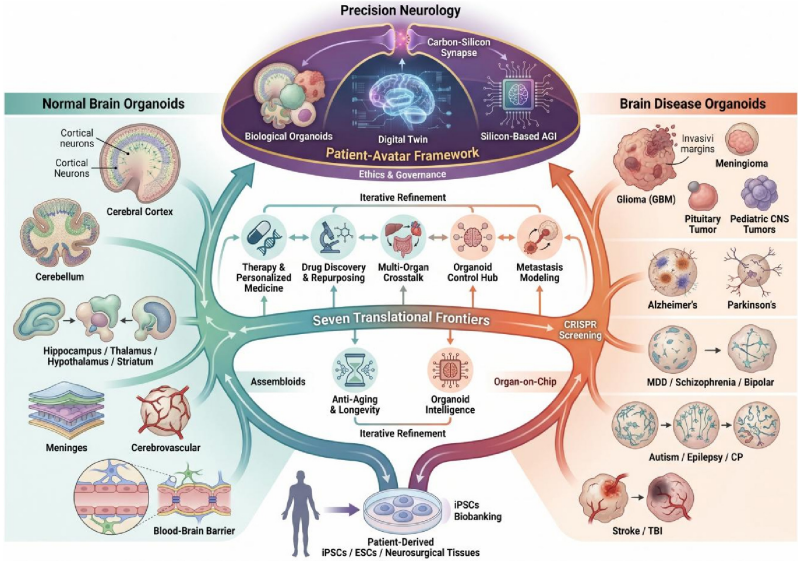
Correspondence

Jianfei Long, longjianfei@huashan.org.cn;
Xinxin Han, xxhan@sibs.ac.cn

In Brief

This comprehensive review charts the translation of human brain organoids from foundational neurodevelopmental models to advanced platforms for disease modeling, therapeutic discovery, and hybrid biocomputing. The authors survey region-specific, vascularized, and neuroimmune systems, critically evaluating preclinical capabilities across central nervous system neoplasms, neurodegenerative diseases, and psychiatric disorders. Finally, the review explores the frontier of organoid intelligence (OI) via the "Patient-Avatar" framework and evaluates the evolving global regulatory landscape, highlighting China's pioneering 2025 national ethical guidelines for high-risk stem cell research.

Graphical abstract



Highlights

- Reference Architecture Defined: Maps the state-of-the-art landscape of normal brain organoids—spanning region-specific identities, barrier systems, and cross-species chimeras—establishing a structural benchmark for pathological modeling.
- Oncology Gaps Illuminated: Covers adult and pediatric neuro-oncology organoid modeling from glioblastoma to midline gliomas, while identifying critical lacunae in understudied entities like vestibular schwannoma.
- Multi-Scale Pathological Modeling: Captures complex multi-cellular and environmental disease dimensions, featuring vascularized neuroimmune models for Alzheimer's and circuit-level assembloids reproducing schizophrenia network defects.
- Regenerative Vascular Reconstruction: Showcases functional angiogenesis breakthroughs where bioinspired matrix-hydrogel systems host vascular organoids to successfully reconstruct ischemic stroke cavities *in vivo*.
- The Patient-Avatar Paradigm: Proposes a tripartite framework integrating patient-derived biology, digital twins, and silicon-based artificial general intelligence (AGI) to pioneer next-generation precision neurology.
- Pioneering Ethical Governance: Analyzes the world's first unified national guidelines (China, 2025) mandating multi-modal consciousness cross-validation, chimera cell-ratio caps, and absolute implantation bans for embryo models.

Cell Organoid

Review

https://doi.org/10.26599/CO.2026.9410024

Brain organoids and brain disease organoids modeling in the age of artificial intelligence

Jianfei Long[1,*] and Xinxin Han[2,3,4,*]

[1] *Department of Pharmacy, Huashan Hospital, Fudan University, Shanghai 200040, China*
[2] *Shanghai LiSheng Biotech, Shanghai 200092, China*
[3] *Organ Regeneration X Lab, LiSheng East China Institute of Biotechnology, Peking University, Nantong 226299, China*
[4] *AGRI-animal organoid institute, South China Agricultural University, Guangzhou 510642, China*

Received: 23 February 2026 / **Revised:** 29 May 2026 / **Accepted:** 4 June 2026

ABSTRACT

The human brain governs bodily functions with exceptional complexity, yet neuroscience research is hindered by limited access to authentic human tissues, restricted availability of region-specific specimens, and a lack of physiologically relevant experimental models. Brain organoids and disease-specific brain organoids have emerged as transformative research platforms to address these bottlenecks. This review systematically summarizes advances in normal brain organoids, including cortical, cerebellar, meningeal, cerebrovascular, and blood–brain barrier models, as well as disease organoids recapitulating glioma, neurodegeneration, psychiatric disorders, neurodevelopmental defects, epilepsy, stroke and other neurological conditions. We highlight seven key translational advances of brain organoids in targeted therapy development, drug discovery and repurposing, brain–organ crosstalk, tumor brain metastasis, and longevity research. We further discuss the frontier interplay between carbon-based brain organoids and silicon-based artificial intelligence. Integrating stem cell biology, tissue engineering and clinical neuroscience, brain organoids greatly advance mechanistic research of neurological diseases and provide promising platforms for personalized medicine and regenerative therapeutics.

KEYWORDS

brain organoid, neurological disease modeling, patient-derived organoid, tumor organoid, personalized medicine, organoid intelligence

Introduction

The human brain's extraordinary complexity—86 billion neurons forming 100 trillion synapses—underpins cognitive capacities that have inspired artificial intelligence development[1,2]. Yet studying this organ directly remains challenging due to ethical constraints and species differences. While rodent models have long dominated neuroscience research, their lissencephalic cortices and shortened developmental timelines poorly replicate the human brain[3,4]. These limitations are compounded by policy restrictions on fetal tissue research, creating an urgent need for alternative human-based platforms[5].

Brain organoids offer a transformative solution. Since the discovery that human pluripotent stem cells can self-organize into cerebral tissues[6], the brain organoid field has rapidly advanced through protocols generating increasingly sophisticated models that capture key aspects of human brain development and organization. A better shift is the use of patient-derived induced pluripotent stem cells (iPSCs) as primary source material[7]. These iPSCs preserve donors' complete genetic backgrounds—including disease-associated variants and epigenetic signatures—enabling the creation of personalized brain organoids that maximize translational relevance[8]. Another major advance is patient-derived glioblastoma organoids, and these patient brain tumor organoids can now be used for cell therapy[1,9,10]. Neurosurgical brain samples, such as glioma tumors, provide additional sources that capture tumor microenvironment complexities[2,11].

Organoids' three-dimensional (3D) systems offer distinct advantages over conventional approaches. Brain organoids maintain human-specific gene expression patterns that rodent models lack[2,12]. They support tissue-level organization that 2D cultures cannot achieve[13]. They enable investigation of disease mechanisms within individual genetic contexts[7]. Their scalability further facilitates high-throughput drug screening applications[14]. Beyond biomedical applications, brain organoids serve as platforms for studying neural computation principles that inform artificial intelligence development[15], potentially illuminating the biological foundations of intelligence itself[16].

*Address correspondence to Jianfei Long, longjianfei@huashan.org.cn; Xinxin Han, xxhan@sibs.ac.cn

Cite this article as Long, J. F., et al. *Cell Organoid*, **2026**, 2: 9410024.

Brain organoid technology overcomes several key barriers. First, humans cannot be used directly as primary research subjects. Second, ethical concerns limit certain studies. Third, there is a growing need for personalized models of CNS development and disease. By using accessible stem cells or brain disease tissues, we can construct brain organoids. These organoids create important opportunities for our time. They help us understand brain development, dysfunction, pathogenesis, and drug screening. They also try to bridge the gap between brain intelligence and artificial intelligence.

In the following sections, we will summarize normal brain organoids and disease-related brain organoids (Fig. 1). Normal brain organoid models include several different types. These cover cerebral cortical maturation, region-specific brain structures, the cerebellum, the meninges, the cerebrovascular and blood–brain barrier systems, and chimeric integration models. Disease-related brain organoids cover many conditions. These include glioblastoma, meningioma, schwannoma, pituitary adenoma, and glioblastoma sarcoma. They also include stroke, brain injury, epilepsy, neurodevelopmental defects, depression, Alzheimer's disease (AD), and Parkinson's syndrome, etc. Together, these brain organoid studies lay a solid foundation and framework. They support research on CNS development and disease pathogenesis. They also guide drug discovery, mechanistic studies, and clinical diagnosis and treatment.

We will now explore the seven key areas where brain organoids are driving progress, one by one. First, brain organoids make personalized treatment and drug development for brain diseases possible—something previously unattainable. Second, brain organoids aid in expanding the indications of existing drugs. This opens a broad door for repurposing old drugs to treat brain diseases. Third, brain organoids help reveal the interactions between the brain and other organs. Fourth, brain organoids can also regulate other organoids. For example, they can be used to study the process of non-brain tumor metastasis to the brain and to develop therapeutic drugs. Fifth, brain organoids also help explore the important role of the brain in anti-aging and healthy longevity. Sixth, we also address an ethical concern regarding brain organoids that need attention.

The seventh and final area is a frontier at the heart of the current wave of artificial intelligence: the relationship between carbon-based neural models—represented by human brain organoids—and silicon-based artificial general intelligence.

Through a visual overview, we will reveal how brain organoids are transforming research on intellectual development and CNS disorders. Brain organoids not only accelerate the development of new therapies, but also dramatically deepen our understanding of brain pathology mechanisms. At the same time, they provide a vital new platform for personalized medicine and regenerative applications—goals that were previously unrealistic.

Normal brain organoids: A baseline for development, prevention, brain function, and disease calibration

Normal, healthy brain organoids serve as a powerful tool for studying brain development and protection, as well as a control group for disease-related brain organoids. Before modeling disorders of the central nervous system, the healthy developmental trajectory of normal brain development must be faithfully recapitulated *in vitro*. Normal brain organoids act as the benchmark for all pathological comparisons, providing an essential baseline for understanding brain disease and brain function. Over the past decade, the field has progressed from generic whole-brain organoids to increasingly sophisticated models representing distinct brain regions and cortical structures. These normal brain organoid models can capture the developmental trajectories, regional identities, and functional integration of specific brain structures. Brain organoids have evolved into practical models for revealing human neurodevelopment, neural circuit assembly, and brain-to-organ communication. We will review recent advances in normal brain organoids, covering a range of normal, healthy models—including those of cerebral cortical maturation, region-specific brain

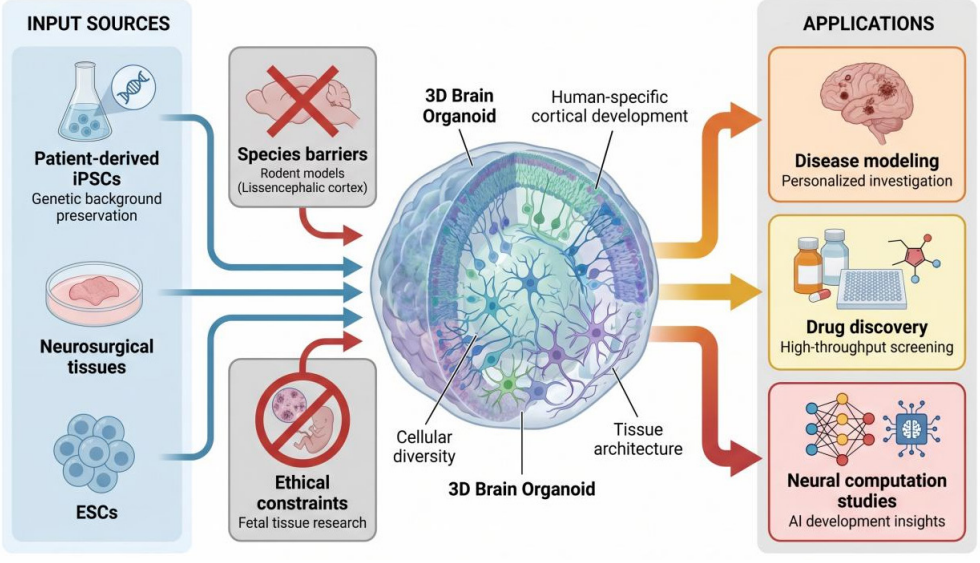


Figure 1. Human brain modeling through organoid technology. Brain organoid technology overcomes key limitations of conventional neuroscience approaches by generating three-dimensional models from patient-derived iPSCs, neurosurgical tissues, and ESCs. These organoids replicate human-specific cortical development, cellular diversity, and tissue architecture. The methodology bypasses species barriers of animal models and ethical constraints on fetal tissue research, while enabling scalable applications in disease modeling, drug discovery, and neural computation studies.

structures, the cerebellum, meninges, cerebrovascular and blood–brain barrier systems, and chimeric brain organoid integration models—as well as the methodological innovations driving the field of normal brain organoids forward.

Cerebral cortex organoids across developmental stages

The cerebral cortex has remained the primary focus of brain organoid research. Lancaster discovered that human pluripotent stem cells can self-organize into cerebral tissues[6]. Early cortical organoids successfully modeled fetal-stage neurogenesis. However, their relevance was initially limited to early human gestation. Gordon and colleagues conducted breakthrough long-term culture work. For example, they cultured cortical organoids for 5, 9, and 12 months. This revealed that long-term culture induces transcriptomic shifts. These shifts match key early postnatal developmental milestones. For instance, at 9–12 months, about 15% of projection neurons aligned with early postnatal stages. At 5 months, only 2% did. Meanwhile, the *in vitro* organoid system also exhibited mature astrocyte signatures and the emergence of oligodendrocyte populations[17]. This finding established that organoids possess an intrinsic developmental clock. This clock can drive molecular maturation without external instructive cues. Thus, it can surpass the fetal period and continue developing.

Faravelli demonstrated unprecedented long-term culture of human brain organoids for up to 5 years. They proved that transcriptomic and epigenetic ages precisely matched *in vitro* culture duration. These ages also paralleled *in vivo* human brain development timelines[18]. Organoids at 3–6 months corresponded to mid-gestation stages. Organoids at 9 months and beyond gradually transitioned toward late prenatal and postnatal equivalence. Strikingly, when neural progenitor cells from these gradually cultured "aged" organoids were transplanted into chimeric organoids, they retained a memory of culture time. They rapidly generated late-born neuronal fates. They also skipped early neuronal progeny production[18]. This observation indicates that progenitor cells within organoids encode temporal information. They execute age-appropriate developmental programs. This provides a revolutionary platform for modeling late developmental events and age-dependent neurological conditions.

As a complement to extended culture approaches, methodological innovations have also improved the spatial organization of cortical organoids. Qian et al. developed sliced human cortical organoids maintained at the air–liquid interface. This enabled sustained expansion of the cortical plate, enhanced neuronal maturation, and the formation of distinct cortical layers[19]. This method allows the generation of both deep-layer (CTIP2+) and upper-layer (SATB2+) excitatory projection neurons. Their proportions more closely resemble *in vivo* corticogenesis[19]. Giandomenico independently demonstrated that air–liquid interface culture of whole cerebral organoids supports the generation of diverse nerve tracts with functional output[20]. Recently, the cortical brain organoid slice (cBOS) method has extended these capabilities. It enables whole-cell patch-clamp recordings, calcium imaging, and metabolic perturbation studies in organized neuronal networks[21].

He et al. integrated a global human neural organoid cell atlas[22]. They collected single-cell transcriptomic data from 36 datasets across 26 laboratories worldwide. Using over 1.7 million single-cell transcriptomic data points from these datasets, they constructed developmental trajectories for neural organoids, including brain organoids. They performed detailed analyses of metabolic features and transcriptomes. This led to the development of HNOCA-tools, a research tool for neurological disorders such as autism and Fragile X syndrome[22]. Hendriks et al., along with their colleagues, developed an *in vitro* brain organoid (FeBO) from human fetal brain tissue. Using CRISPR-Cas9 technology, they found that introducing TP53 gene defects into a small number of cells within these brain organoids caused these TP53-deficient cells to rapidly outcompete others[23]. Hendriks also used CRISPR-Cas9 technology to attempt to turn off glioblastoma-related genes TP53, PTEN, and NF1. They then observed the changes in drug response[23].

In summary, these advances in long-term maturation and structural refinement have elevated cortical organoids from fetal neurodevelopmental models to a platform. This platform can be used to investigate postnatal circuit maturation, early-life neurological transitions, and brain disease modeling, diagnosis, and treatment research.

Region-specific brain organoids: beyond the cortex

The cerebral cortex has drawn much attention. Yet many subcortical structures also govern critical neurological functions. Their organoid models have now matured considerably.

Midbrain organoids containing functional dopaminergic neurons have become robust platforms for Parkinson's disease (PD) research[24]. Smits et al. showed that midbrain-like organoids derived from patient iPSCs with LRRK2 mutations exhibit fewer dopaminergic neurons and abnormal neuromelanin production. This established a model for studying dopaminergic neurodegeneration[24]. More recent protocols have further refined dopaminergic neuron subtype specification and integration capacity[25].

Hippocampal organoids represent another key advance. Sakaguchi et al. generated functional hippocampal neurons from self-organizing human embryonic stem cell-derived dorsomedial telencephalic tissue. They used precisely controlled BMP and Wnt signaling gradients to induce medial pallium identity[26]. These organoids produced both Zbtb20+/Prox1+ granule-like neurons and Zbtb20+/KA1+ pyramidal-like neurons. Both subtypes exhibited electrophysiological functionality and network-forming capacity[26]. Recent work further integrated hippocampal organoids with three-dimensional liquid metal-based neuro-interfaces. This enabled high-throughput electrophysiological recording across 128 channels and detection of glutamate-mediated oscillatory network activity[27].

Thalamic organoids have opened new avenues for modeling sensory information relay circuits. Xiang et al. established human embryonic stem cell-derived thalamic organoids (hThOs). They used single-cell RNA sequencing to characterize thalamic lineage formation and its divergence from telencephalic fates[28]. Critically, when fused with cortical organoids, these thalamic organoids established reciprocal projections. This created a thalamocortical assembly system capable of modeling bidirectional circuit connectivity[28]. Duenki et al. recently extended this work. They constructed multi-organoid loop cerebral connectoids linking thalamic and cortical organoids. These connectoids demonstrated enhanced neural network dynamics and sequence-specific entrainment[29]. Patton and colleagues further showed that thalamocortical assembloids exhibit canonical synaptic plasticity—including long-term potentiation and depression. This represents the first demonstration of such plasticity rules in a fully human *in vitro* system[30].

Hypothalamic organoids, particularly those modeling the arcuate nucleus, have advanced metabolic neuroscience. Huang et al. generated hypothalamic arcuate organoids from human iPSCs. These organoids contained functional POMC and AgRP neurons capable of responding to metabolic hormones, including leptin, insulin, and ghrelin[31]. These organoids provide a human-specific platform for investigating the neural circuitry of energy balance. They have implications for obesity and diabetes research[31]. Adjacent pituitary organoid protocols have also matured. Recent single-cell transcriptomic and spatial omics analyses have revealed the cellular diversity and SOX3-dependent developmental mechanisms underlying human pituitary organogenesis[32].

Striatal organoids have been successfully generated and integrated with cortical spheroids to form cortico-striatal assembloids. This enables the study of human interneuron migration and long-range circuit assembly[33]. Choroid plexus organoids spontaneously form fluid-filled cysts producing cerebrospinal fluid-like material. They offer complementary platforms for studying blood–cerebrospinal fluid barrier function and central nervous system drug penetration[34].

Budinger et al. applied multidimensional technologies, including single-cell transcriptomic sequencing and spatial transcriptomics[35]. For the first time, they comprehensively mapped the spatiotemporal landscape of human fetal midbrain development from early to mid-gestation. They then compared the developmental trajectory and atlas of *in vitro* midbrain organoids with those of the *in vivo* midbrain. Using patient-derived organoids from dopamine transporter deficiency syndrome (DTDS), they further uncovered the pathogenesis of DTDS[35]. This work provides a new three-dimensional atlas for midbrain development research and neurological disease modeling. Wang et al. developed an interesting "Large Organoid-Cutting-Small Piece-Repopulation" strategy to generate DA-neuron-rich midbrain organoids from hESCs[36]. These organoids enable long-term functional testing and serve as a PD model.

Collectively, these region-specific organoid models have transformed brain organoid research from a cortex-centric system into a multi-regional platform. This platform can model the distributed circuitry underlying complex brain functions.

Cerebellar organoids

The cerebellum, essential for motor coordination, cognitive processing, and language function, presents distinct developmental challenges due to its protracted developmental timeline and specialized cellular architecture. Muguruma et al. established the foundational protocol for cerebellar organoid generation, demonstrating that human pluripotent stem cells could self-organize into polarized cerebellar tissue containing both granule cell progenitors and Purkinje cells in three-dimensional culture[37]. This protocol recapitulated key aspects of cerebellar development, including the generation of neurons from two distinct germinal zones—the rhombic lip and the ventricular zone—establishing a framework for subsequent refinements.

A major recent advance came from Atamian et al., who reported the generation of human cerebellar organoids with functionally mature Purkinje cells[38]. Through optimized differentiation protocols incorporating TGFβ inhibition combined with FGF2 and insulin treatment, the authors produced cerebellar organoids containing rhombic lip-derived granule cell progenitors and ventricular zone-derived Purkinje cells. Long-term cultured organoids developed spontaneous neuronal activity, and whole-cell patch-clamp recordings confirmed the functional maturation of Purkinje cells, including the emergence of complex firing patterns characteristic of this principal cerebellar neuron type[38]. Parallel work has enabled cell type-specific visualization of FOXP2+ Purkinje cells through CRISPR-mediated reporter knock-in, facilitating live-cell imaging and the isolation of this population for downstream analysis[39].

Several methodological improvements have enhanced the scalability and maturity of cerebellar organoid systems. Chen et al. reported optimized long-term culture conditions that support extended Purkinje cell survival and maturation[40], while Silva and colleagues developed bioreactor-based protocols for scalable production of homogeneous cerebellar organoids[41]. Nayler et al. further demonstrated that air–liquid interface culture improves cerebellar organoid viability and maturation, enabling single-cell transcriptomic profiling of cerebellar cell types[42]. Despite these advances, significant challenges persist. Cerebellar organoids currently lack the precise spatial lamination of the mature cerebellar cortex—the molecular layer, Purkinje cell layer, and granule cell layer—and the long-term survival of Purkinje cells remains suboptimal compared with *in vivo* counterparts. The formation of complete cerebellar circuit motifs, including climbing fiber and mossy fiber inputs, awaits further methodological innovation.

Meningeal organoids

Meningeal organoid research is still sparse, but some clues exist[43]. Human cortical organoids (hCOs) typically lack the surrounding meningeal layer, which is essential for normal corticogenesis and brain development. Jalilian and Shin first generated hCOs from the single rosettes. Using a 3D co-culture system, they then encapsulated brain organoids with a thin layer of meningeal cells from very early stages of cortical development[43]. This strategy aimed to mimic the *in vivo* cortical brain structure, including meningeal cells, more closely.

Despite scattered clues, systematic protocols for meningeal organoid culture and modeling remain absent. The meninges—three membranous layers (dura mater, arachnoid mater, and pia mater)—enveloping the brain and spinal cord—play vital roles in brain development, homeostasis, and immune surveillance. Yet, meningeal organoid modeling remains a conspicuous gap. This void reflects several intertwined challenges: the cellular complexity of meningeal tissue, including fibroblasts, macrophages, vascular endothelial cells, and specialized barrier-forming cells; the lack of well-characterized, meninges-specific molecular markers to purify and validate meningeal cell types; and the unclear developmental origins of the meningeal layers, which arise from multiple embryonic sources, including the neural crest and mesoderm. Moreover, the meninges establish critical signaling gradients—secreting retinoic acid, Wingless/INT-related proteins, and chemokines to regulate cortical neurogenesis, neuronal migration, and axon guidance. This implies that any competent meningeal organoid must recapitulate not only cellular composition but also these instructive paracrine functions.

Certain brain organoid systems reportedly contain meningeal-like cells as incidental populations. Pellegrini et al., in their CNS barrier organoids, noted the presence of fibroblast-related cells mapping to pial and vascular reference data[34]. Some transplantation studies have also observed meningeal-like tissue formation at the organoid–host interface[44]. However, these incidental findings do not constitute validated meningeal organoid

models. Potential routes for future development include: adapting meningeal fibroblast culture protocols to three-dimensional conditions; utilizing iPSC-derived mesenchymal lineages to generate pial cell models; employing defined morphogen combinations to direct neural crest or mesodermal progenitors toward meningeal fates; and meningioma organoids in disease states carrying meningeal cells. Co-culture systems combining meningeal cell precursors with brain organoids may offer an intermediate strategy to study brain–meninges interactions without forming fully self-organized meningeal organoids. Establishing meningeal organoids would be especially valuable for studying meningeal roles in cerebrospinal fluid dynamics, neuroimmune surveillance, and the pathogenesis of meningiomas and other meningeal disorders, representing a critical frontier.

Cerebrovascular and blood–brain barrier organoids

Traditional brain organoids largely lack all functional vasculature of the brain, such as blood vessels, lymphatic vessels, and the choroid plexus. This absence has long been a key limitation, restricting studies of nutrient delivery, oxygenation, and neurovascular interactions. Multiple strategies have emerged to address this challenge. Co-culture approaches have proven effective: Shi et al. developed vascularized human cortical organoids (vOrganoids) by integrating human umbilical vein endothelial cells into developing organoids. These endothelial cells formed vessel-like structures in ventricular-zone-like regions, reducing apoptosis while increasing the proportion of spontaneously firing neurons[45]. Cakir et al. used a genetic engineering strategy, demonstrating that ectopic ETV2 transcription factor expression in iPSC-derived organoids induced vessel-like networks with blood–brain barrier features, including tight junction formation and polarized expression of barrier transporters[46].

Transplantation-based and integration-based vascularization offers another viable route to vascularize brain organoids. Mansour et al. demonstrated that human brain organoids transplanted into the mouse brain became vascularized by host-derived endothelial cells, forming functional vessels with blood–brain barrier properties[44]. This *in vivo* vascularization strategy not only attempts to resolve nutrient limitation but also enables studies of neurovascular coupling and BBB development in a physiologically relevant context. The engineering of intact neurovascular units has advanced, integrating endothelial cells, pericytes, astrocytes, neurons, and microglia into three-dimensional neurovascular models[47,48]. A 3D human neurovascular model has been established specifically designed for AD research, integrating multiple neurovascular unit cell types into brain organoids[47,49].

Choroid plexus organoids represent a complementary approach to modeling CNS barrier function. Pellegrini et al. demonstrated that choroid plexus organoids form tight epithelial barriers, secrete CSF-like fluid, and accurately predict CNS drug permeability—for example, correctly distinguishing impermeable dopamine from L-dopa, which enters via LAT1-mediated transport[34]. Air–liquid interface culture methods, including the cBOS approach, further enhance functional maturation of vascularized organoid systems by improving oxygen delivery and enabling long-term maintenance of complex neural networks[21]. Together, these vascularization strategies are progressively transforming avascular brain organoids into systems capable of modeling neurovascular disease, BBB drug transport, and cerebrovascular developmental disorders.

Chimeric brain organoids and cross-species integration

Transplanting human brain organoids into animal hosts both promotes maturation beyond what is achievable *in vitro* and enables assessment of neural circuit integration potential. The host brain environment provides vascularization, sensory input, and neuromodulatory signals absent in standard culture conditions, thereby accelerating neuronal maturation and enabling functional circuit integration. Revah et al. demonstrated that human cortical organoids transplanted into the somatosensory cortex of newborn rats matured extensively, developing morphologically complex neurons with elaborate dendritic arbors and functional synapses[47]. Single-nucleus transcriptomic analysis revealed ongoing corticogenesis and the emergence of activity-dependent transcriptional programs in transplanted organoids. Functionally, transplanted human neurons received thalamocortical and cortico-cortical inputs, responded to sensory stimuli, and their optogenetic activation drove reward-seeking behavior in host animals—demonstrating genuine functional integration with host sensory and motivational circuits[47].

Jgamadze et al. extended these findings by transplanting human forebrain organoids into the injured adult rat visual cortex and demonstrating structural and functional integration with the host visual system[50]. Transplanted neurons established afferent and efferent connections, received polysynaptic input from the host retina, and exhibited orientation-selective responses to visual stimuli[50]. Dong et al. further showed that human brain organoids transplanted into the mouse brain can establish subcortical projections, including long-range connections essential for functional neural circuit repair[51]. Collectively, these transplantation studies demonstrate that human organoid-derived neurons can integrate into host neural circuits across species boundaries, supporting their potential utility in neural repair applications.

Transplantation into non-human primate models raises the possibility of clinical translation. Kitahara et al. reported that human brain organoids transplanted into the primary motor cortex of cynomolgus macaques survived for at least 12 weeks without overgrowth, integrated with the host vasculature, and sent axonal projections along the corticospinal tract to the corpus callosum, striatum, and adjacent cortical regions[52]. The use of non-human primate hosts addresses important issues of species-specific developmental timing and immunological responses that rodent models cannot fully replicate. These chimeric approaches raise significant ethical considerations concerning the creation of human–animal neural chimeras, including concerns about potential alterations to animal cognition or behavior. These ethical dimensions require careful oversight and will be discussed in detail in subsequent sections. Nonetheless, cross-species transplantation represents a critical bridge between *in vitro* organoid modeling and potential therapeutic applications in human patients.

Methodological advances in normal brain organoid construction

Beyond biological advances in regional specification and maturation, methodological innovations have improved the reproducibility, scalability, and experimental utility of brain organoid systems. A fundamental challenge of traditional stem-cell-derived brain organoid protocols is the stochastic formation of multiple independent neuroepithelial rosettes within each

organoid, each rosette acting as a separate organizing center, resulting in structural heterogeneity. Knight et al. addressed this limitation by developing self-organizing single-rossette (SOSR) organoids, in which precise control of starting cell density and matrix composition ensures that each organoid contains exactly one neural rosette[53]. These SOSR organoids exhibit predictable size, cytoarchitecture, and developmental trajectories, markedly improving batch-to-batch reproducibility and making them especially suitable for quantitative experimental comparisons and high-throughput screening applications[53].

The integration of CRISPR-based genetic screening with brain organoid technology has, in parallel, opened transformative possibilities for systematic genetic studies. Li et al. established the CHOOSE system (CRISPR-human organoids-single-cell RNA sequencing), enabling pooled loss-of-function screening of 36 high-risk autism spectrum disorder genes in mosaic forebrain organoids, with single-cell transcriptomics as the readout[54]. This approach revealed cell-type-specific developmental defects, identifying dorsal intermediate progenitors and upper-layer excitatory neurons as the cell populations most vulnerable to Autism spectrum disorder (ASD)-associated gene disruptions[54]. Recent protocol standardization has extended these capabilities: Nature Protocols now provides detailed workflows for conducting arrayed CRISPR interference and CRISPR activation screens in human neural organoids and assembloids, lowering technical barriers for widespread adoption[55]. Complementary work from Harvard researchers applied arrayed CRISPRi screening to identify transcriptional regulators controlling anterior neural tube closure—including ZIC2, SOX11, and ZNF521 —linking specific gene perturbations to morphogenetic outcomes in human neural development[56]. These screening platforms have also elucidated multi-scale mechanisms underlying species differences in brain development, where comparative CRISPR interference screens have revealed how CDK2- and CCNE1-mediated cell cycle dependencies drive distinctive neural progenitor expansion between the human and ape lineages[57].

A particularly innovative advance involves the reconstruction of the ventricular–subventricular zone (V-SVZ) neurogenic niche. Vassalou et al. demonstrated that overexpressing GEMC1 or MCIDAS transcription factors in human brain organoids drives radial glial cells toward an ependymal cell fate, generating multiciliated ependymal cells that assemble into characteristic pinwheel-like structures together with GFAP+ astrocytes[58]. This represents the first successful *in vitro* reconstruction of the human adult V-SVZ niche, providing a platform to study adult neurogenesis, ependymal cell biology, and the mechanisms by which the CSF–neural stem cell interface regulates brain homeostasis[58]. These technological innovations—single-rosette organoid generation, CRISPR-based genetic screening, and niche reconstruction—collectively accelerate the transformation of brain organoid technology from a specialized methodology into a standardized platform accessible to the broader neuroscience community.

Brain disease organoids I: Brain tumor organoids

Primary CNS tumors display extraordinary diversity. Each tumor type varies between patients, exhibiting pronounced heterogeneity not only across tumor types but also among individuals with the same diagnosis. This disease and patient heterogeneity manifest in cellular origins, molecular drivers, clinical behavior, and therapeutic responsiveness. Brain tumor organoid technology has emerged as an innovative platform for modeling these diverse malignancies, striving to faithfully recapitulate tumor architecture, cellular heterogeneity, and microenvironment interactions that traditional 2D culture and animal models cannot capture. Patient-derived tumor organoids preserve the genetic landscape and histopathological features of the parent tumor while supporting experimental manipulations infeasible *in vivo*. We outline the current status of organoid models spanning the full spectrum of CNS tumors, from the most intensively studied adult gliomas to rare pediatric tumors, evaluating established models and identifying tumor types for which no organoid systems have yet been reported. We further describe existing alternative preclinical approaches for these gaps, providing a detailed analysis of significant advances and remaining voids in the brain tumor field.

Glioblastoma and high-grade glioma organoids

Among all brain tumor types, glioblastoma (GBM; WHO grade 4) has received the most extensive attention in organoid research. This reflects both its clinical urgency as the most common and lethal primary malignant brain tumor in adults and its inherent biological complexity. Jacob et al. established the foundational patient-derived glioblastoma organoid (GBO) biobank, generating organoids from 53 patient samples that faithfully reproduced inter- and intra-tumoral heterogeneity, preserved cancer stem cell populations, maintained hypoxic gradients, and retained tumor microenvironment features[11]. These GBOs subsequently enabled systematic pharmacogenomic studies across large patient cohorts, revealing shared and patient-specific therapeutic vulnerabilities linked to genomic and transcriptomic profiles.

Recently, Logun et al. demonstrated that patient-derived GBOs can serve as real-time avatars for evaluating clinical CAR-T cell therapy responses, showing that organoid responses correlated with actual patient MRI responses and CSF cytokine profiles, thereby establishing proof-of-principle for organoid-guided personalized immunotherapy[59]. Baisiwala et al. further advanced this paradigm by developing the integrated human organoid tumor–immune (iHOTT) model, an autologous co-culture system combining patient-derived GBM cells with matched peripheral blood mononuclear cells within human cortical organoids. This system recapitulated patient-specific immune responses, T cell infiltration patterns, and cytokine profiles, with pembrolizumab treatment mirroring clinical cell-type shifts and T cell receptor sequencing revealing drug-responsive clonotype expansion[10].

Brain organoid–glioma co-culture systems represent a complementary approach, in which patient-derived glioma cells or spheres are implanted into healthy brain organoids, enabling the study of tumor invasion within a physiologically relevant neural microenvironment[60,61]. These systems have revealed that GBM cells hijack neural progenitor cell migration pathways for invasion and remodel the organoid microenvironment through paracrine signaling[61]. Genetically engineered brain organoid tumor models, in which oncogenic mutations are introduced in developing organoids, enable the study of tumor initiation and early transformation events inaccessible in patient-derived samples[62]. Ge et al. used organoid tumor transplantation to discover PTPRZ1-mediated glioblastoma–microenvironment communication, demonstrating that reduced brain-derived PTPRZ1 paradoxically increases tumor invasiveness through non-enzymatic signaling pathways[63]. The GlioME model developed by

Zheng et al. represents another advance for preserving the intact glioma microenvironment in personalized drug screening[64].

Beyond conventional GBM, the rare biphasic variant gliosarcoma was recently modeled through patient-derived organoids established by Park et al. They established gliosarcoma organoids (GSOs) from freshly resected tumors with 100% success. These GSOs retained characteristic glial and mesenchymal differentiation, stromal cell diversity, and case-specific somatic variants. Single-cell RNA sequencing revealed enrichment of fibroblast-like and oligodendrocyte precursor cell-like states, distinguishing them from GBM organoids[65]. The pioneering 3D GBM organoid culture established by Hubert et al. demonstrated that organoid culture maintains hypoxic gradients and cancer stem cell heterogeneity not preserved in 2D systems[66]—a finding that underlies much subsequent GBO research.

Low-grade and intermediate-grade glioma organoids

For low-grade diffuse gliomas, the organoid modeling landscape remains far less developed than for GBM, reflecting slower growth kinetics, lower culture success rates, and unique microenvironmental requirements. Patient-derived organoids from IDH-mutant astrocytomas (WHO grades 2–4) have been successfully established, preserving key metabolic features, including 2-hydroxyglutarate accumulation, IDH1 R132H mutant expression, and characteristic histological features[9]. Organoids derived from diffuse astrocytoma (WHO grade 2) and anaplastic astrocytoma (WHO grade 3) maintain cytoarchitecture, stemness markers, and vascular endothelial features; however, grade 2 samples exhibit lower 3D growth capacity and lower overall success rates compared to high-grade counterparts[9]. Kim et al. documented serial progression from grade 2 to grade 4 in the same patient over an 11-year clinical course, with corresponding longitudinal cell and tumor sphere models successfully capturing the molecular and functional facets of malignant transformation at each stage[67].

For oligodendrogliomas, IDH-mutant and 1p/19q-deleted (WHO grades 2–3), organoid modeling has proven particularly challenging due to their characteristically slow growth and tight dependence on neuronal synaptic connections. A breakthrough study by Raleigh et al. (2025) established patient-derived organoid co-cultures of oligodendroglioma with cortical neurons, demonstrating that recurrent oligodendrogliomas enrich synaptic gene expression programs and that neuronal hyperexcitability in organoid co-cultures drives tumor proliferation. In this system, pharmacological blockade of neurophysiological signaling pathways suppressed oligodendroglioma growth[68]. Scaffold-based 3D culture systems have also been applied to oligodendrogliomas with moderate success[69].

Diffuse midline glioma, H3K27-altered (formerly diffuse intrinsic pontine glioma), constitutes a distinct entity where the field has achieved notable organoid progress. A landmark 2025 study established de novo H3.3K27M-altered diffuse midline glioma organoids (DMGOs) in human brainstem organoids using region-specific organoid engineering, recapitulating the pontine-specific developmental context of this universally fatal pediatric tumor[70]. These DMGOs closely resembled primary patient material by single-cell RNA sequencing, displaying OPC-like, AC-like, and MES-like tumor cell states, with global methylation profiles clustering with human DMG rather than GBM or posterior fossa ependymoma[70]. Earlier work by Haag et al. demonstrated that H3.3-K27M drives neural-stem-cell-specific gliomagenesis in human iPSC-derived models[71], while Bressan et al. showed that the regional identity of neural stem cells determines their oncogenic response to H3.3 mutants[72], together establishing the critical importance of developmental context in DMG organoid modeling.

For pilocytic astrocytoma (WHO grade 1), the most common pediatric low-grade glioma, no organoid-based model has been reported to date. This tumor type typically harbors KIAA1549:BRAF fusions or BRAF V600E mutations. Research has utilized 2D cell lines (including DKFZ-BT66), 3D sphere cultures, genetically engineered mouse models carrying BRAF-KIAA fusions, and patient-derived xenografts. Similarly, no organoid model has been established for subependymal giant cell astrocytoma (SEGA), a WHO grade 1 tumor associated with tuberous sclerosis complex and mTOR pathway hyperactivation. Although TSC-derived human brain organoids have been developed to model cortical tuber formation, these models model developmental lesions rather than SEGA tumors themselves. For both entities, establishing reliable organoid systems represents a major unmet need, which would facilitate preclinical testing of MAPK pathway and mTOR inhibitors, respectively.

Meningioma organoids

Meningioma, the most common primary CNS tumor in adults, has long lacked reliable *in vitro* models that recapitulate its diverse molecular subtypes, variable clinical behavior, and distinctive microenvironment composition. Recent advances have filled this gap through the establishment of patient-derived meningioma organoids by two independent groups using complementary approaches. Zohdy et al. established meningioma organoids (MEN-Os) from 15 surgical samples encompassing WHO grades 1 and 2, demonstrating retention of histological features, meningioma-specific markers (EMA, progesterone receptor), and transcriptional signatures including NF2, CDKN2A, and TP53 expression[73]. Deconvolution analysis confirmed preservation of tumor, stromal, and immune microenvironment components, with organoids successfully maintained for up to four weeks[73].

Concurrently, Kim et al. developed microenvironment-preserving meningioma organoids (MNOs) from four patients using Matrigel-free liquid culture. These organoids maintained native histology and the tumor microenvironment without enzymatic dissociation, were successfully maintained for over nine weeks with cryopreservation capability, and demonstrated the anti-tumor efficacy of mifepristone[74]. Together, these platforms enable the study of meningioma biology across the full spectrum from benign to atypical variants and support drug screening for recurrent or refractory cases. Both studies demonstrated successful establishment from NF2-mutant meningiomas, addressing one of the most common molecular subtypes. However, organoid models for anaplastic (WHO grade 3) meningioma, which carries the poorest prognosis, and for tumors bearing clinically targetable TRAF7, AKT1, and SMO mutations, remain to be established to enable targeted therapy testing.

Vestibular schwannoma organoids

To date, no organoid model has been established for vestibular schwannoma (acoustic neuroma). This tumor type arises from the vestibular branch of the eighth cranial nerve and is associated with NF2 gene mutations in both neurofibromatosis type 2-related

schwannomatosis and sporadic cases. Its slow growth, biphasic Antoni A and Antoni B histological patterns, and the lack of optimized culture conditions for vestibular-nerve-derived Schwann cells render it exceptionally difficult to culture *in vitro*. Available research models include iPSC-derived Merlin-deficient Schwann-cell-like spheroids, which recapitulate molecular alterations found in patient tumors and have been used to test antisense oligonucleotide-based therapies[75]; the HEI193 immortalized vestibular schwannoma cell line; NF2 knockout genetically engineered mouse models; patient-derived xenografts; and primary cell cultures with only short-term viability. The absence of authentic patient-derived vestibular schwannoma organoids represents a critical gap, especially given that no FDA-approved pharmacotherapy currently exists for NF2-related schwannomatosis and there is a pressing clinical need for preclinical platforms to test hearing preservation strategies and targeted agents such as bevacizumab.

Pituitary tumor organoids

Pituitary adenomas (pituitary neuroendocrine tumors, PitNETs) have been modeled on a limited scale with organoid technology, with most progress centered on corticotroph adenomas causing Cushing's disease. Chakrabarti et al. established the first authentic patient-derived pituitary adenoma organoids (hPITOs) from transsphenoidal surgical samples of Cushing's disease patients. These organoids replicated the cellular complexity and endocrine function of parent tumors and demonstrated patient-specific drug responses through high-throughput screening[76]. Zhang et al. had previously developed human ACTH-secreting tumor-like models with organoid features, providing an early platform for studying corticotroph tumor biology[77]. Complementing patient-derived approaches, genetically engineered pituitary organoids carrying USP48 or USP8 mutations have been developed from iPSCs to model corticotroph tumorigenesis, with these models showing differential responses to glucocorticoid receptor modulators, including relacorilant and mifepristone[78]. Organoid models for the remaining pituitary adenoma subtypes—including somatotroph (GH-secreting), lactotroph (PRL-secreting), thyrotroph, gonadotroph, and null cell adenomas—remain very limited or unpublished, representing a substantial gap given the distinct molecular drivers and clinical management challenges of each subtype.

Craniopharyngioma, a sellar region tumor divided into adamantinomatous (pediatric, CTNNB1-mutant) and papillary (adult, BRAF V600E-mutant) subtypes, currently has no established organoid models. Research relies on primary cell cultures, which progressively lose epithelial phenotype and keratinizing features with extended passaging[79], and genetically engineered mouse models[80]. The distinctive histological features of craniopharyngioma—including wet keratin, palisading epithelium, and calcification—coupled with its intimate anatomical relationship to the hypothalamus, pose unique challenges that organoid culture has yet to overcome.

Pediatric and other CNS tumor organoids

Pediatric brain tumors differ fundamentally from adult malignancies in cellular origins, molecular drivers, and therapeutic responsiveness. Their organoid modeling has advanced substantially in recent years. Medulloblastoma, the most common malignant pediatric brain tumor, has been modeled through both genetically engineered cerebellar organoids and patient-derived approaches. Ballabio et al. demonstrated that Otx2 and c-MYC overexpression in human cerebellar organoids generates medulloblastoma-like organoids whose DNA methylation profiles cluster with human Group 3 tumors, while SMARCA4 overexpression or the EZH2 inhibitor tazemetostat reduces tumorigenesis[81]. The same team subsequently showed that Notch1 signaling determines the competence of cerebellar organoid progenitors for SHH-type versus Group 3/4 medulloblastoma induction[82]. Lago et al. have established a comprehensive protocol suite for medulloblastoma organoid generation, drug screening, lineage tracing, co-culture, and *in vivo* experiments[83], facilitating broader adoption of these models. Medulloblastoma organoids have been generated from fresh surgical tissue using a universal growth-factor-free medium through the Children's Brain Tumor Network (CBTN) platform, with successful biobanking and revival for precision medicine testing, including CAR-T therapy[84]. All four molecular subgroups—SHH, WNT, Group 3, and Group 4—have been represented in organoid models, although Group 3 and Group 4 model establishment remains more difficult than for SHH-driven tumors.

Ependymoma organoid models have been developed primarily through the CBTN platform for pediatric cases, which has generated over 30 organoid models covering posterior fossa (PFA and PFB) and supratentorial ependymomas[84]. These organoids demonstrate phenotypic representation of clinical histology and maintain stable growth following biobanking. For atypical teratoid/rhabdoid tumor (ATRT), a highly aggressive infantile embryonal tumor characterized by SMARCB1 or SMARCA4 loss, patient-derived tumoroid models were established in 2023 for the MYC and SHH molecular subgroups. These tumoroids preserved the molecular and phenotypic features of parent tumors with minimal drift during long-term culture and revealed subgroup-specific drug vulnerabilities[85]. Notably, the ATRT-TYR subgroup has not been successfully cultured as organoids, mirroring the difficulty in establishing patient-derived xenograft models for this subtype.

Primary CNS lymphoma (PCNSL), an aggressive non-Hodgkin lymphoma confined to the CNS, had its first organoid model established by Li et al. in 2025. These CNS lymphoma organoids (CLOs) were generated from patient samples within two weeks and accurately modeled the histological features, gene expression profiles, mutational spectra, and cell-type heterogeneity of original tumors. Single-nucleus RNA sequencing confirmed the preservation of molecular signatures, and therapeutic profiling revealed specific sensitivity to methotrexate[86].

Several important CNS tumor types currently lack any reported organoid models. Pineoblastoma, a rare and aggressive pineal embryonal tumor often associated with RB1 pathway alterations, has no organoid model, with research limited to rare cell lines, genetically engineered mouse models, and limited patient-derived xenografts. CNS germ cell tumors, including germinomas, teratomas, embryonal carcinomas, yolk sac tumors, and choriocarcinomas, similarly lack organoid-based models, a gap likely reflecting the rarity of these tumors and the limited availability of patient tissue for research. Ganglioglioma, the most common epilepsy-associated glioneuronal tumor, has also not been modeled through organoid technology. For each of these entities, patient-derived xenografts, rare established cell lines, and genetically engineered mouse models remain the primary

preclinical tools available. Systematic expansion of organoid biobanking initiatives, particularly those focused on rare pediatric tumors, is essential to address these longstanding gaps in preclinical research.

Brain disease organoids II: Neurodegenerative, vascular, psychiatric, and developmental disorders

The previous section outlined progress in modeling normal brain and specific disease organoids, such as brain tumors. However, the scope of brain disease organoid research extends well beyond these paradigms, encompassing an increasingly broad range of neurodegenerative, cerebrovascular, neurodevelopmental, and psychiatric disorders. The past two years have witnessed particularly rapid expansion, with the emergence of vascularized neuroimmune organoids for AD, large-scale neurodevelopmental disorder biobanks, T cell and microglial co-culture systems for PD, and AI-integrated organoid platforms for psychiatric disease classification. What follows is a comprehensive overview of these developments, organized by disease category, highlighting both established models and the latest breakthroughs from 2025–2026. For diseases lacking dedicated organoid models, we explicitly note this limitation and describe the most relevant alternative approaches, ensuring an accurate assessment of the field's current capabilities.

Alzheimer's disease

Brain organoid models of AD have progressed from recapitulating single pathological features to integrating multiple disease-relevant cell types and environmental insults within unified experimental systems. Early work by Gonzalez et al. demonstrated that brain organoids derived from iPSCs of familial AD patients—or engineered to carry amyloid precursor protein (APP) and presenilin 1 (PSEN1) mutations—generate amyloid-β aggregates and hyperphosphorylated tau pathology[87]. Subsequent studies identified that APOE4, the strongest genetic risk factor for sporadic AD, causes widespread molecular perturbations in brain organoids, including impaired myelination due to oligodendrocyte cholesterol dysregulation—directly linking the major genetic susceptibility locus to white matter pathology[88]. Recent advances in multicellular and environmentally stressed organoid systems have substantially expanded these foundational observations.

Several 2025–2026 advances have significantly broadened the translational relevance of AD organoid models. Serotonergic hindbrain organoids generated from AD patient iPSCs have been used to evaluate antidepressant drug responses at the molecular level, with escitalopram treatment partially restoring serotonin signaling and cell–cell communication proteins in responder-derived organoids[89]. Notably, inter-individual differences in drug response were readily observable, highlighting the value of patient-derived organoids in predicting individual treatment outcomes. Extracellular vesicles (EVs) secreted from AD patient-derived organoids have emerged as promising liquid biopsy biomarkers, with reduced RAB3A, NSF, and ATCAY protein levels in EVs reflecting disrupted neuronal signaling pathways. These vesicular cargo profiles may serve as minimally invasive indicators of disease staging and drug response[89].

The pathogen infection hypothesis of AD has gained substantial support from organoid studies: human brain organoids infected with herpes simplex virus type 1 (HSV-1) recapitulated multi-scale AD neuropathology, including amyloid-β deposition, reactive gliosis, neuroinflammation, and neuronal loss, with antiviral drugs such as ribavirin and valacyclovir partially rescuing these phenotypes[90].

Complementing these findings, 2026 reports demonstrated that HIV-1 infection induces neuroinflammatory cascades and synaptic dysfunction in brain organoids, further validating the infectious etiology theory through human-relevant *in vitro* systems. Environmental pollutant exposure and gut–brain axis perturbations have also been modeled in AD organoid systems, with these environmental dimensions formally recognized through a dedicated funding initiative by the National Institute of Environmental Health Sciences in 2025. For sporadic AD without known causative mutations, serum-exposed organoid culture has proven effective, modeling blood–brain barrier disruption as an early pathological event and implicating peripheral factors in disease initiation[91].

Collectively, these advances position vascularized, microglia-integrated, and environmentally stressed organoid systems as increasingly powerful platforms for mechanistic discovery and therapeutic screening in AD.

Parkinson's disease

PD organoid research is built upon midbrain-specific differentiation protocols that generate functional dopaminergic neurons, the selectively vulnerable population in the disease. Smits et al. established the foundational method for generating midbrain-like organoids from iPSCs of PD patients carrying LRRK2 mutations, observing reduced dopaminergic neuron numbers and aberrant neuromelanin production[24]. These models subsequently enabled detailed investigation of α-synuclein aggregation, mitochondrial dysfunction, and selective dopaminergic neuron vulnerability—the core pathogenic features of PD[92]. Jo et al. further demonstrated that PD patient iPSC-derived midbrain organoids form neuromelanin-like granules and undergo age-dependent dopaminergic neuron degeneration, recapitulating the temporal dimension of disease progression that requires extended culture periods to manifest[93]. The 3D architecture of organoids provides unique advantages for PD modeling, supporting the study of both cell-autonomous and non-cell-autonomous mechanisms, including astrocytic and microglial contributions to neurodegeneration.

Recent advances have introduced innovative co-culture systems and more refined mechanistic insights. A T cell–midbrain organoid co-culture model reported in 2025 enables direct investigation of neuroimmune interactions in PD, providing a platform to explore T cell-mediated neurodegenerative mechanisms[94]. Microglia-specific contributions have been elucidated through an LRRK2-G2019S microglia–organoid co-culture system: mutant microglia adopted a pro-inflammatory phenotype characterized by elevated TNFα secretion, selectively inducing loss of TH-positive dopaminergic neurons in co-cultured midbrain organoids. Notably, glycolytic inhibition with oxamate rescued dopaminergic neuron survival, defining a metabolically driven neuroinflammatory axis[95].

Single-cell transcriptomic analysis of LRRK2-G2019S midbrain organoids has further revealed disease-associated transcriptional signatures and altered developmental trajectories[96]. Mitochondrial studies in 2025 demonstrated that axonal mitochondria of dopaminergic neurons exhibit characteristically shorter length and

lower membrane potential—features that may underlie their selective vulnerability in PD. PRKN mutations attenuated these mitochondrial differences between dopaminergic and non-dopaminergic neurons, suggesting broader effects on cellular bioenergetics[97]. Complementing these cellular models, gut–brain axis studies have linked gut-derived metabolites to dopaminergic neuron function and neuroinflammatory modulation, with multi-organ systems connecting the midbrain, liver, and gut organoids demonstrating that gut-derived factors significantly regulate dopaminergic pathways[98]. Together, these advances establish PD organoid research as a multi-system endeavor encompassing neuronal intrinsic vulnerability, glial–neuroimmune crosstalk, and peripheral organ interactions.

Major depressive disorder and psychiatric disorders

Psychiatric disorders pose unique modeling challenges due to their polygenic architecture, environmental influences, and the difficulty of recapitulating behavioral phenotypes *in vitro*. Nevertheless, brain organoids and iPSC-derived neural models have made substantial contributions, particularly for major depressive disorder (MDD), bipolar disorder (BD), and schizophrenia. For MDD, Vadodaria et al. identified serotonin-induced hyperactivity associated with SSRI treatment resistance in patient-derived neurons, providing a cellular correlate of pharmacological non-response[99]. Recent 2025 work extended this framework, demonstrating that esketamine—an NMDA receptor antagonist approved for treatment-resistant depression—induces distinct electrophysiological and metabolic reprogramming effects in brain organoids, illuminating rapid-acting antidepressant mechanisms[100]. Studies of MDD with suicidal behavior using ventral forebrain organoids further revealed reduced 5-HT2C serotonin receptor expression in GABAergic interneurons, accompanied by altered neurite branching and calcium signaling[101].

Bipolar disorder organoid research achieved a major advance in 2025 with the first comprehensive analysis of mitochondrial and inflammatory vulnerabilities in patient-derived brain organoids. El Soufi El Sabbagh et al. demonstrated that BD patient-derived organoids exhibit mitochondrial dysfunction, metabolic dysregulation, and heightened sensitivity to NLRP3 inflammasome activation. Pharmacological inhibition with the selective NLRP3 inhibitor MCC950 effectively rescued mitochondrial function and reduced inflammatory activation, establishing the mitochondrial–inflammasome axis as a core pathophysiological mechanism in BD[102]. Complementing these molecular studies, machine learning algorithms applied to iPSC-derived neural models have distinguished electrophysiological signatures of BD with increasing accuracy, achieving 83% disease classification accuracy, rising to 92% after electrical stimulation[103]. The BD2 network has subsequently launched an iPSC and organoid working group to establish standardized cell repositories and optimize quality control protocols, reflecting growing institutional investment in BD organoid research[102].

Schizophrenia organoid models have undergone particularly rapid refinement. Building on earlier observations of transcriptomic alterations in synaptic biology and neurodevelopment[104], along with ventricular neuropathology accompanied by disrupted neurogenesis[105], 2025 witnessed a transformative advance: Walsh et al. developed forebrain assembloid systems capable of generating PVALB-positive cortical interneurons within 120 days. These interneurons matched primary PVALB interneurons in molecular and electrophysiological properties, and schizophrenia-associated structural variants disrupted their migration, molecular identity, and ability to modulate cortical network activity, including gamma oscillations[106]. This work provided the first functional demonstration of interneuron-mediated gamma oscillation deficits in an entirely human *in vitro* system, bridging a critical gap between genetic risk and circuit-level dysfunction. AI-based classification has further sharpened diagnostic precision: support vector machine classifiers applied to cortical interneuron cultures achieved 95.8% accuracy in distinguishing schizophrenia, while brain-like activity patterns from organoids served as disease biomarkers with 83% accuracy at baseline and 92% after stimulation[103].

Multi-omics analyses have continued to identify schizophrenia risk factors associated with GABAergic pathway dysregulation, with PCCB emerging as a key gene linked to tricarboxylic acid cycle disruption and reduced GABA levels in forebrain organoids[107]. Proteomic analyses of schizophrenia-derived iPSC-derived neural cells and brain organoids have confirmed molecular signatures consistent with postmortem brain tissue, validating the fidelity of organoids as reliable proxies for the diseased human brain[108].

For anxiety disorders and obsessive-compulsive disorder (OCD), no dedicated brain organoid models currently exist. The neural circuit basis of these disorders—including anxiety-associated amygdala–hippocampal–prefrontal connectivity and OCD-related cortico-striatal-thalamo-cortical circuitry—exceeds the structural complexity achievable with current organoid technology. Furthermore, the absence of clear monogenic etiologies and the inability to measure anxiety-like or compulsive behaviors *in vitro* have limited model development. Research in these areas relies primarily on 2D iPSC neuronal cultures and animal models. Although iPSC lines from OCD patients have been established, functional characterization in published studies has not been performed[109]. As organoid technology advances toward region-specific assembloids capable of recapitulating elements of limbic and frontostriatal circuitry, opportunities to model anxiety disorders and OCD may emerge.

Developmental brain disorders

Brain organoids have proven exceptionally powerful for modeling neurodevelopmental disorders, as they recapitulate the sequential processes of neural induction, progenitor expansion, neuronal migration, and cortical layer formation that characterize human embryogenesis. Lancaster et al. established this capability through their seminal demonstration that CDK5RAP2-mutant organoids exhibited premature neuronal differentiation and reduced progenitor pools, directly linking radial glial division disruptions to primary microcephaly pathogenesis[6].

The Zika virus epidemic accelerated methodological development: multiple groups demonstrated that the virus selectively infects neural progenitors in organoids, triggering cell death, reduced organoid size, and disrupted cortical layering—phenotypes mirroring the microcephaly observed in infected infants[110]. These foundational observations have been substantially extended through systematic genetic studies and large-scale biobanking initiatives.

The most significant advance in developmental brain disorder modeling in 2025 came from the Gleeson team's establishment of a large-scale neurodevelopmental disorder (NDD) organoid

biobank. This resource contains 352 patient-derived iPSC lines representing microcephaly, polymicrogyria, epilepsy, and intellectual disability, from which over 6,000 brain organoids were generated and characterized by histology and single-cell transcriptomics[111]. Analysis revealed disease-category-specific convergent phenotypes: microcephaly organoids showed cell survival defects with excess TTR-positive cells; polymicrogyria organoids exhibited intermediate progenitor junctional defects; epilepsy organoids displayed excessive astrogliosis; and intellectual disability organoids showed overproduction of TTR-positive cells. This biobank represents the largest systematic NDD organoid resource to date, providing an unprecedented foundation for comparative disease mechanism studies.

Complementing the biobank approach, 2025 gene-specific studies identified novel microcephaly genes—including NARS1, whose biallelic mutations reduce radial glial proliferation in cortical organoids, and RTF1/ASAP2, validated through CRISPR-Cas9 knockout in neural progenitors and organoids[112,113]. For cortical malformations, organoid models have proven effective in dissecting cell cycle and cytoskeletal dynamics underlying global malformations, and mTOR pathway alterations in focal cortical dysplasia[113].

Cerebral palsy merits specific mention as a condition without a direct organoid model. Cerebral palsy encompasses a heterogeneous group of permanent movement disorders arising from non-progressive insults to the developing fetal or infant brain. Its multifactorial etiology—including prematurity, perinatal hypoxic-ischemic injury, intracranial hemorrhage, and genetic variants—has hindered the development of dedicated cerebral palsy organoid systems. Instead, researchers have employed hypoxic-ischemic injury models in brain organoids as mechanistic surrogates, exposing organoids to low-oxygen conditions to model the energy failure, excitotoxicity, and neuroinflammation characteristic of perinatal brain injury[114]. A 2025 study using fused vascularized brain organoids demonstrated that vascularized systems mount a stronger inflammatory response to hypoxic injury while exhibiting greater progenitor resilience, suggesting that vascular elements significantly modulate hypoxic-ischemic outcomes[115]. Sex-specific differences were also observed, with female organoids activating survival pathways under hypoxic stress, while male organoids exhibited chaperone system dysregulation and protein misfolding stress[116]. These findings, while not directly modeling cerebral palsy, provide mechanistic insights into the hypoxic-ischemic injury that constitutes the major etiology of the condition.

Autism spectrum disorder

ASD organoid research has progressed from descriptive phenotyping to systematic genetic screening and convergent mechanism identification. Paulsen et al. established foundational observations by identifying shared developmental phenotypes across organoids from different ASD patients, including accelerated neuronal differentiation and desynchronized GABAergic interneuron development—findings that reframe ASD as a disorder rooted in disrupted developmental trajectories rather than mature circuit dysfunction[117].

Li et al. introduced the CHOOSE system (CRISPR-human organoids-single-cell RNA sequencing), a landmark study enabling pooled loss-of-function screening of 36 high-risk ASD genes in mosaic brain organoids. This work identified dorsal intermediate progenitors and upper-layer excitatory neurons as the cell types most vulnerable to ASD-associated gene perturbations, revealing that diverse genetic risk factors converge on specific developmental cell populations[54]. Complementary Perturb-seq studies have systematically characterized cell fate regulatory networks, illuminating how regulatory gene networks govern human cortical development and how their disruption leads to ASD-associated phenotypes[118].

Recent advances have extended this framework in two important directions. First, myelination defects have emerged as a convergent feature of ASD organoid models. ASD genetic risk factors are known to converge on neural stem cells and oligodendrocyte precursor cells (OPCs), affecting their proliferation and differentiation. Both ASD organoid models and postmortem tissue exhibit dysregulated myelin-related transcriptional signatures, while pro-myelinating drugs have shown therapeutic potential in complementary animal models[119].

These observations connect ASD to white matter abnormalities previously identified through neuroimaging, providing a cellular and molecular foundation for therapeutic strategies targeting myelination. Second, the convergence of developmental desynchronization across multiple ASD risk genes has been further validated. Large-cohort single-cell genomic analyses have revealed ASD-associated molecular cascades and cell-type-specific alterations, with excitation–inhibition imbalance emerging as a consistent theme across diverse genetic etiologies[120].

The NDD organoid biobank has provided independent large-scale validation of ASD-specific cellular defects, while MEF2C-associated ASD has been modeled in patient-derived neurons and organoids, revealing microRNA dysregulation and excitation–inhibition imbalance[121]. Together, these advances position ASD organoid research at the forefront of developmental neurobiology, with the CHOOSE system and its successors enabling systematic dissection of polygenic risk at single-cell resolution.

Epilepsy

Epilepsy organoid models have evolved from demonstrating aberrant electrophysiological activity to serving as functional drug screening platforms for specific genetic epilepsy syndromes. Samarasinghe et al. established foundational capability by identifying spontaneous neural oscillations and epileptiform activity in organoids from patients with genetic epilepsy syndromes, with aberrant network activity—including high-frequency oscillations and ictal-like events—modulated by antiseizure medications[122]. These observations provided the first proof-of-principle that organoid systems can recapitulate the hyperexcitable network states characteristic of epilepsy. WWOX-deficient organoid studies further elucidated epileptic encephalopathy mechanisms, revealing disrupted neuronal differentiation and hyperexcitability, linking genetic loss-of-function to electrical phenotypes[123].

The 2025–2026 period has brought significant technical and translational advances. For SCN2A-associated epilepsy, a microfluidic multi-organoid drug screening platform utilizing 3D microelectrode arrays (3D-MEA) was developed, enabling simultaneous recordings from multiple organoids under different pharmacological conditions[124].

SCN2A-mutant epilepsy organoids exhibited significantly higher firing and bursting rates compared to control organoids, and the platform successfully identified carbamazepine and clobazam as effective suppressors of aberrant discharges—demonstrating the feasibility of patient-specific antiseizure drug

screening in an organoid high-throughput format. Independent studies showed that 4-aminopyridine (4-AP) reliably induces epileptiform activity in human stem-cell-derived neural organoids, characterized by increased spike frequency, bursting, and synchrony.

Three mechanistically distinct antiseizure medications—carbamazepine, ethosuximide, and diazepam—each eliminated these epileptiform patterns, validating organoids as responsive systems for antiseizure drug discovery[125].

These platforms complement ongoing Dravet syndrome research, where SCN1A mutations affecting inhibitory interneuron function have been modeled in patient-derived excitatory and inhibitory cortical spheroids[126], and observations from the large-scale NDD biobank that epilepsy organoids consistently display excessive astrogliosis as a convergent cellular phenotype[111].

Together, these developments position epilepsy as one of the most translationally mature application areas for brain organoid technology, with direct relevance to precision pharmacology for genetically defined epilepsy syndromes.

Stroke and traumatic brain injury

Stroke and traumatic brain injury (TBI) represent acute neurological injuries for which organoid models have progressed from modeling initial injury mechanisms to exploring regenerative therapeutic strategies. For ischemic stroke, hypoxic treatment of brain organoids has been the primary modeling approach, recapitulating the cell death, inflammation, and tissue damage characteristic of cerebral ischemia[114]. Zhu et al. extended this framework by developing a multi-omics stroke organoid platform that integrates hypoxic injury with transcriptomic, proteomic, and metabolomic analyses, identifying astrocyte subpopulations with distinct hypoxic responses and discovering candidate therapeutic compounds[127]. These models enable testing of neuroprotective strategies in a human cellular context, addressing a critical translational gap, as many neuroprotective candidates successful in rodent stroke models have failed in human clinical trials.

A defining 2025 advance in stroke organoid research came from the development of a non-expansile biodegradable matrix (NEBM) hydrogel system for vascular organoid transplantation in ischemic stroke. Xiao et al. from Army Medical University demonstrated that NEBM—a biomimetic hydrogel providing anti-swelling support while permitting growth—can accommodate blood vessel organoids (BVOs) that develop organ-specific vascular networks and functionally anastomose with host vasculature when transplanted into stroke cavities[128]. This approach addresses a fundamental limitation of stroke recovery: the progressive clearance of extracellular matrix from the ischemic cavity prevents cellular infiltration and regenerative processes. The NEBM-BVO system achieved functional vascular reconstruction within the infarct region, with transplanted animals demonstrating motor function improvement at two weeks and progressive angiogenesis and axonal ingrowth at twelve weeks. This study represents the first demonstration of vascular organoid-based therapeutic angiogenesis in stroke and provides proof-of-concept for organoid transplantation as a regenerative strategy for ischemic brain injury.

For traumatic brain injury, organoid models have proven valuable in dissecting the molecular sequelae of mechanical neurotrauma. A 2025 study using isogenic 3D cortical organoids carrying MAPT mutations demonstrated that mechanical compression injury induced tau hyperphosphorylation and oligomerization in wild-type human organoids, while the V337M mutation exacerbated cellular damage—revealing synergistic gene–environment interactions between genetic tauopathy risk and traumatic injury[129]. Complementary work using high-intensity ultrasound platforms identified KCNJ2, a mechanosensitive potassium channel, as a therapeutic target in TBI. Genome-wide CRISPR interference screening in injured organoids demonstrated that KCNJ2 inhibition mitigates neurodegeneration both *in vitro* and *in vivo*, while simultaneously identifying TDP-43 proteinopathy as a key driver of cell death following mechanical injury[130]. Together, these stroke and TBI models demonstrate that organoid technology can address both the acute pathophysiology and long-term regenerative challenges of cerebrovascular and traumatic neurological injuries.

Emerging disease applications

Beyond established disease categories, brain organoid technology is being applied to an expanding repertoire of rare and emerging neurological disorders, as well as fundamental neuropharmacology and neurotoxicology platforms. A major institutional advance in 2026 was the launch of the NIH Drug Research Organoid Integrated Development Platform (DROIDp), which integrates 3D brain organoids, multi-electrode array electrophysiology, and artificial intelligence to create a standardized national resource for neuropharmacology and neurotoxicology screening[131]. The DROIDp platform aims to provide high-throughput, human-relevant preclinical evaluation of CNS drug candidates and environmental toxicants, addressing the long-standing translational gap between animal models and human neural responses. This initiative signals growing recognition at the federal funding level that brain organoids represent essential infrastructure for neuropharmacological drug development.

For rare monogenic neurological disorders, organoid models of SYNGAP1-related intellectual disability have revealed an excitation–inhibition imbalance and synaptic dysfunction, complementing broader NDD biobanking efforts[111]. Monogenic studies continue to expand the catalog of rare diseases amenable to organoid modeling, with each new model reinforcing the finding that diverse genetic etiologies frequently converge on shared cellular pathways—particularly excitation–inhibition balance, synaptic function, and neuroimmune modulation. The neurological complications of SARS-CoV-2, initially modeled through brain organoid infection studies demonstrating preferential infection of specific neuronal populations, including dopaminergic neurons[132], have now been extended to show that viral infection triggers premature senescence and distinct inflammatory signatures—phenotypes mitigated by senolytic treatment[133]. These findings continue to inform understanding of COVID-19-associated cognitive dysfunction and accelerated neurodegeneration risk. As organoid technology continues to mature, the convergence of patient-derived modeling, CRISPR-based genetic engineering, multi-omics profiling, and AI-driven analysis positions brain organoids increasingly as central tools for both elucidating rare disease mechanisms and conducting large-scale therapeutic discovery across the full spectrum of neurological disorders.

Clinical translation of brain organoids

Translating brain organoid technology from basic research to clinical application represents a critical frontier in precision neurology. Although brain organoids remain primarily employed as research tools, multiple clinically relevant avenues have emerged with the potential to reshape the diagnosis and treatment of neurological disorders.

Patient-derived organoids for personalized medicine

Brain organoids derived from patient-specific induced pluripotent stem cells (iPSCs) have opened new avenues for personalized medicine, enabling therapeutic strategies tailored to an individual's genetic background. In neuro-oncology, patient-derived glioblastoma organoids have been used for *in vitro* drug sensitivity testing, helping identify the most effective treatment regimens for individual patients[11]. The creation of glioblastoma organoid biobanks has supported systematic pharmacogenomic studies across large patient cohorts, revealing both shared and patient-specific therapeutic vulnerabilities[62].

Beyond oncology, patient-derived organoids have been employed to characterize the functional effects of rare genetic variants identified through clinical whole-genome sequencing. They establish mechanistic links between genotype and neurological phenotype, supporting clinical decision-making. The concept of living biobanks composed of patient-derived brain organoids is gaining momentum: systematically generating, characterizing, and sharing organoids from diverse patient populations for collaborative research accelerates target discovery across different genetic and demographic backgrounds[134].

In vivo transplantation and circuit integration

A landmark step in the clinical translation of brain organoids came from the study by Revah et al. Human cortical organoids transplanted into the somatosensory cortex of newborn rats matured extensively, formed morphologically complex neurons and functional synapses, and integrated into host sensory and reward circuits[47]. Transplanted human neurons received thalamocortical input, responded to sensory stimuli, and their optogenetic activation triggered reward-seeking behavior in host animals.

This work provided proof-of-principle for using organoid-derived neural tissue for brain repair while simultaneously raising important ethical questions about cross-species neural chimeras. Dong et al. further extended this approach by demonstrating that human brain organoids transplanted into the mouse brain can form subcortical projections, proving the long-range connectivity necessary for functional neural circuit repair[51]. These results support organoid transplantation as a potential future therapy for focal brain injuries, including stroke, traumatic brain injury, and neurodegenerative diseases.

Standardization and quality control

Clinical application of brain organoids necessitates rigorous standardization of generation protocols and quality control measures. Methods to reduce batch-to-batch variability have improved the reproducibility of cortical organoid production[135,136]. Furthermore, a unified nomenclature for neural organoids and assembloids has been established to improve communication within the field[136].

Cellular stress artifacts, particularly those caused by limited nutrient and oxygen supply in the organoid core, have been recognized and mitigated through slicing and air–liquid interface culture[53,55]. Single-cell transcriptomic profiling is now routinely employed for quality control, allowing precise comparison of organoid cellular composition with human brain tissue reference data[4,137]. Recent comprehensive assessments of brain organoid technology have developed frameworks to evaluate fidelity across multiple dimensions, including cellular diversity, spatial architecture, and functional maturation[137].

Drug innovation

The pharmaceutical industry has long been plagued by high attrition rates in central nervous system (CNS) drug development. Over 90% of clinical trials for neurological disorders end in failure, a rate substantially higher than in other therapeutic areas. Much of this failure stems from the limited predictive power of traditional preclinical models, particularly rodent systems, which often fail to faithfully recapitulate human brain physiology and pharmacology. Brain organoids have emerged as transformative platforms for CNS drug discovery, screening, and repurposing.

Drug discovery and high-throughput screening

The scalability of brain organoid culture in multi-well plates supports the establishment of high-throughput screening (HTS) platforms for CNS drug candidates[138]. Organoid-based assays integrate multiple functional and molecular readouts, such as calcium imaging for neural activity, multi-electrode array recordings for network electrophysiology, and single-cell transcriptomics for molecular profiling. These collectively provide a more comprehensive pharmacological profile than traditional cell-based assays.

Brain organoid HTS has also accelerated drug repurposing by discovering novel neuroprotective or neurotrophic activities among already-approved drugs, enabling faster translation to the clinic by leveraging existing safety data[139]. For example, senolytic screening in aged brain organoids has identified dasatinib, quercetin, and fisetin as agents that selectively eliminate senescent neural cells and restore youthful transcriptomic profiles[133].

Blood–brain barrier and neurotoxicity models

The blood–brain barrier (BBB) remains a major obstacle to effective CNS drug delivery. Traditional *in vitro* BBB models rely primarily on endothelial cell monolayers and struggle to replicate the 3D multicellular architecture of the human neurovascular unit. Brain organoids engineered to include endothelial cells and pericytes can now model BBB permeability and drug transport more realistically within a 3D microenvironment[46,140].

Complementing these systems, choroid plexus organoids spontaneously form fluid-filled cysts that produce CSF-like fluid, providing a novel platform to study drug entry across the blood–CSF barrier[34].

Brain organoids also offer distinct advantages in neurotoxicity testing. They allow assessment, in a species-relevant manner, of how developing neural tissue responds to toxicants, drugs, and infectious agents. For instance, organoid models have been used to demonstrate that SARS-CoV-2 preferentially infects specific neuronal populations, including dopaminergic neurons[132], with direct implications for understanding COVID-19-associated

neurological complications and developing protective interventions.

Accelerating the drug development pipeline

Incorporating brain organoids into the drug pipeline can reduce the cost and time of CNS drug development. By generating human-relevant efficacy and toxicity data early in preclinical testing, organoid screening helps prioritize candidates with greater clinical potential and reduce late-stage trial failures.

Panels of patient-derived organoids further allow drug responses to be stratified according to genetic background, supporting patient enrichment strategies in clinical trials and advancing precision pharmacology for neurological diseases[141]. Recent work integrating organoid intelligence with developmental neurotoxicity testing has further broadened the application scope of brain organoids in drug screening[142].

Brain–x axis: Multi-organ interactions

The brain does not operate in isolation. Bidirectional communication between the brain and peripheral organs through neural, endocrine, immune, and metabolic pathways plays a crucial role in both physiological homeostasis and disease pathogenesis. When combined with multi-organ assembloids and organ-on-chip systems, brain organoid technology enables researchers to systematically dissect these brain–peripheral organ axes in human-relevant cellular models.

The gut–brain axis

The gut–brain axis, describing bidirectional signaling between the CNS, the enteric nervous system, the gut microbiome, and the intestinal epithelium, has emerged as a major focus in modern neuroscience[143]. Disrupted signaling along this axis is implicated in diverse disorders, including PD, autism spectrum disorder, depression, and anxiety.

Workman et al. first established a foundational *in vitro* platform by generating intestinal organoids containing functional enteric neurons, enabling direct modeling of gut–brain interactions[144]. More recently, co-culture systems combining brain and gut organoids have been used to study how microbial metabolites, such as short-chain fatty acids and tryptophan derivatives, influence neural activity and inflammatory responses in human neural tissue.

Trapecar et al. developed a biomimetic multi-organ system connecting human brain, liver, and gut organoids through circulating medium, demonstrating that gut-derived metabolites strongly modulate dopaminergic neuron function and neuroinflammatory pathways[98]. These insights advance our understanding of the microbiome's role in neurological disorders and support the design of microbiome-targeted therapeutics.

Assembloid technology for multi-organ integration

Assembloids—structures formed by fusing organoids of different brain regions or different organ systems—have dramatically expanded the capabilities of organoid research. Birey et al. pioneered this approach by fusing dorsal cortical and ventral ganglionic eminence forebrain spheroids, enabling the study of human interneuron migration *in vitro*[145].

Subsequent assembloid models have integrated cortical–striatal[33], cortical–motor[146], and cortical–spinal modules, generating increasingly complex systems for studying neural circuit assembly and function. Patton and colleagues recently demonstrated synaptic plasticity in human thalamocortical assembloids, showing that thalamic and cortical organoids form reciprocal connections capable of long-term potentiation and depression—the first demonstration of classic synaptic plasticity rules in an entirely human *in vitro* system[30].

The assembloid strategy has also been extended beyond the nervous system to create brain–immune, brain–vascular, and brain–endocrine interfaces, each revealing how peripheral tissues influence neural development and disease[8].

Multi-organ-on-chip platforms

Microfluidic multi-organ-on-chip systems offer a complementary strategy for modeling brain–organ interactions, enabling studies in tightly controlled microenvironments with defined fluid flow, mechanical cues, and structured inter-compartment signaling.

Skardal et al. described an integrated three-tissue organ-on-chip platform connecting liver, heart, and brain organoids through a common circulatory loop, supporting studies of systemic drug effects and inter-organ toxicity[147]. Such systems have revealed that drug-induced liver injury can trigger secondary neurotoxicity through the release of hepatocyte-derived metabolites, underscoring the importance of considering systemic pharmacology in CNS drug development.

As multi-organ platforms grow increasingly sophisticated—some now integrating ten or more organ systems—they promise to provide unprecedented insights into the systemic interactions governing neurological health and disease[148].

Brain health mechanisms

Brain organoids offer unique insights not only into disease mechanisms but also into the fundamental processes that maintain normal brain function and health. The capacity to observe neural circuit assembly, myelination, immune surveillance, and vascular integration in real time within human cellular systems deepens our understanding of the biological foundations of brain health.

Functional neural circuits and oscillatory dynamics

The formation of functional neural circuits serves as a key benchmark for the physiological validity of brain organoids. Sharf and colleagues used high-density CMOS microelectrode arrays and shank electrodes to map spontaneous extracellular activity, revealing structured network connectivity characterized by a core of strong connections amidst widespread weak connections—an architecture resembling *in vivo* cortical networks[149].

Trujillo et al. further demonstrated that cortical organoids generate complex oscillatory patterns, including nested gamma oscillations, which recapitulate the electrophysiological maturation of the preterm human brain[150]. Together, these findings establish brain organoids as viable systems for studying the developmental emergence of neural computation and how network dysfunction contributes to neurological disease.

Analysis of chromatin accessibility during forebrain organoid development has also revealed the epigenetic landscape governing the sequential generation of neuronal subtypes, providing a molecular framework for understanding circuit assembly[151].

Myelination and white matter integrity

Myelination, the process by which oligodendrocytes wrap axons

with lipid-rich myelin to support rapid saltatory conduction, is essential for healthy brain function and is disrupted in multiple sclerosis, schizophrenia, and other disorders.

Madhavan and colleagues successfully induced myelinating oligodendrocytes in human cortical spheroids, showing that these cells form compact myelin sheaths around axons in 3D culture[152]. Marton and colleagues extended these observations, demonstrating that the differentiation and maturation of oligodendrocytes in 3D neural cultures closely mirrors the timeline of myelination *in vivo* in humans[153].

Such organoid models have revealed that APOE4-driven oligodendrocyte cholesterol dysregulation impairs myelination, establishing a mechanistic link between the strongest genetic risk factor for AD and white matter pathology[88].

Neuroimmune interactions

The integration of microglia—the brain's resident immune cells—into brain organoids has enabled detailed investigation of neuroimmune interactions critical to brain health.

Ormel and colleagues discovered that microglia develop innately within brain organoids under specific culture conditions, exhibiting the morphological and transcriptomic signatures of homeostatic brain microglia[154]. Schafer and colleagues constructed an *in vivo* neuroimmune organoid model by implanting microglia-containing organoids into immunocompetent mice, demonstrating that human microglia maintain species-specific transcriptomic signatures, survey the tissue environment, and respond to local injury and systemic inflammation[155].

Park and colleagues further revealed that iPSC-derived microglia support brain organoid maturation through cholesterol transfer, uncovering a previously unrecognized neurotrophic role in neurodevelopment[156].

Vascularization and the neurovascular unit

A major limitation of traditional brain organoids has been the absence of a functional vascular system. Multiple strategies now address this challenge, including co-culture with endothelial cells[157], genetic engineering to induce vessel-like structures[46], and *in vivo* transplantation, where host tissue vascularizes the transplanted organoid[44].

These vascularized models support studies of BBB formation, the coupling of angiogenesis and neurogenesis, and the role of the neurovascular unit in brain homeostasis. Microfluidic systems incorporating brain extracellular matrix have also enhanced the structural and functional maturation of brain organoids, indicating that biomechanical signals from the vascular microenvironment shape neural development[140].

Studies integrating astrocyte maturation with organoid platforms have further highlighted the essential role of astrocytes in assembling and maintaining the neurovascular unit[158].

Inter-organ pathology and tumor metastasis

Many neurological disorders arise from pathological interactions between the brain and peripheral organ systems. Brain metastasis, systemic inflammation, and tumor microenvironment remodeling are key inter-organ processes now amenable to dissection with brain organoid models at unprecedented resolution.

Modeling brain metastasis

Brain metastasis, affecting 20%–40% of patients with systemic cancers, represents a major clinical challenge with limited effective therapies. Choe et al. established brain organoid models of brain metastasis by co-culturing extracranial cancer cells with human embryonic stem cell-derived brain organoids. They demonstrated that metastatic cells actively invade the organoid parenchyma and form expanding colonies[159].

These systems reveal that the brain organoid microenvironment, including astrocytes and microglia, potently modulates metastatic cell survival, proliferation, and therapeutic resistance. Organoid-based BBB models further permit investigation of metastatic cell extravasation into the brain—a critical step in the metastatic cascade difficult to recapitulate in traditional *in vitro* assays.

Glioblastoma–brain microenvironment interactions

Interactions between glioblastoma and its host brain microenvironment are now known to shape therapeutic resistance and disease recurrence. Pine and colleagues showed that the native tumor microenvironment sustains the cellular states observed in primary glioblastoma. These phenotypes are lost in standard 2D culture but preserved in organoid co-culture systems[160].

Krieger et al. combined human brain organoid–glioblastoma co-cultures with single-cell transcriptomics to map tumor invasion at high resolution. They identified distinct transcriptional programs in invading versus proliferating glioblastoma populations[61]. These results support the development of therapies targeting the tumor–brain interface, rather than tumor cells alone.

Neuroinflammation as Inter-Organ Pathology

Systemic inflammation, triggered by infection, autoimmunity, or metabolic disorders, can severely disrupt brain function by compromising BBB integrity, activating microglia, and injuring neurons. Microglia-integrated brain organoid models support investigation of human-specific neuroinflammatory cascades[155,161].

Infection of brain organoids with SARS-CoV-2 induces cellular senescence and distinct inflammatory signatures, providing a mechanistic explanation for COVID-19-associated neurological complications such as cognitive dysfunction and accelerated neurodegeneration[133,134]. Such models are invaluable for elucidating how peripheral infection and systemic immune activation drive CNS pathology, increasingly recognized as a significant contributor to AD, depression, and other chronic conditions.

Longevity neuroscience: Brain aging and regeneration

The brain is one of the organs most profoundly affected by aging. Age-related cognitive decline, neurodegeneration, and cerebrovascular damage profoundly impact quality of life in the elderly. Brain organoids have emerged as powerful tools for uncovering the molecular and cellular mechanisms of brain aging and for testing interventions aimed at extending cognitive healthspan.

Cellular senescence in brain organoids

Cellular senescence—irreversible cell cycle arrest accompanied by a pro-inflammatory secretory phenotype—is a key driver of brain aging and neurodegeneration[161]. Aguado et al. made a landmark discovery: senescent cells gradually accumulate in long-term cultured brain organoids. Treatment of these organoids with senolytic drugs (dasatinib plus quercetin, or fisetin) selectively eliminated senescent cells, reduced age-related inflammation, and reversed transcriptomic aging clocks[133].

The same team extended this work to COVID-19, showing that SARS-CoV-2 infection triggers premature senescence in brain organoids—an effect blocked by senolytic treatment. This finding provides both mechanistic understanding and therapeutic potential for virus-induced neurological aging. Clearance of senescent glial cells prevents tau-dependent pathology and cognitive decline in mouse models[162], and brain organoids now allow researchers to test whether similar senolytic strategies are effective in human neural tissue. Notably, brain organoids can be cultured long-term—for up to several years—offering a unique opportunity to observe the temporal dynamics of cellular senescence, including early biomarkers preceding overt pathological changes.

Epigenetic aging and regeneration

Epigenetic aging clocks—tools that estimate biological age using DNA methylation patterns—have been applied to brain organoids to track age-related epigenomic changes under controlled *in vitro* conditions. Long-term cultured brain organoids undergo progressive epigenetic aging, correlating with transcriptomic alterations typical of the aging human brain: upregulation of inflammatory pathways and downregulation of synaptic and mitochondrial genes[17].

This molecular aging trajectory provides a foundation for testing epigenetic rejuvenation strategies, such as partial reprogramming approaches that reset cellular age without altering cell identity. The expanding hallmarks of aging—genomic instability, telomere attrition, epigenetic alterations, loss of proteostasis, deregulated nutrient sensing, mitochondrial dysfunction, cellular senescence, stem cell exhaustion, and altered intercellular communication—provide a comprehensive framework for evaluating anti-aging interventions in brain organoids[161].

Neuroprotective strategies

Researchers have used brain organoids to test various neuroprotective compounds and interventions, including antioxidants, anti-inflammatory agents, neurotrophic factors, and metabolic modulators. A key advantage of organoid-based neuroprotection studies is the ability to evaluate drug effects in physiologically relevant 3D human neural tissue, with readouts encompassing cell viability, neural network function, and transcriptomic profiling. Identifying compounds that preserve neural function in aging organoids can accelerate the development of interventions that promote healthy human brain aging[91].

The future of hybrid intelligence: Where carbon meets silicon

One of the most provocative and forward-looking applications of brain organoid technology lies in its integration with artificial intelligence and biological computing. The emerging field of "organoid intelligence" (OI) posits that biological neural networks—with their energy efficiency, parallel processing capacity, and robust learning ability—can be harnessed to develop hybrid bio-electronic computing systems. These systems may complement or even surpass traditional silicon-based architectures. Through the "Patient-Avatar-Surrogate" concept, this integration heralds innovative and imaginative visions for how biological intelligence and artificial general intelligence (AGI) may interact. In the AI era, with humanity possessing more leisure time and energy, might there be greater creativity and pursuit of the cosmos

Organoid intelligence

Researchers at Johns Hopkins University formally established organoid intelligence as a distinct scientific discipline through a series of publications. Smirnova, Hartung, and colleagues proposed that human brain organoids, which replicate the cellular diversity and synaptic connectivity of the human brain, could serve as the basis for a novel form of biological computing[16]. The Baltimore Declaration, issued following the first Organoid Intelligence Workshop in 2022, urged the international scientific community to explore the potential of brain organoids for biocomputing while simultaneously establishing ethical guidelines for such research[163]. The remarkable computational efficiency of the human brain inspired this concept: it performs complex cognitive tasks using only about 20 watts of power, vastly outperforming comparable AI systems consuming megawatts of energy.

DishBrain and biological computing

Scientists at Cortical Labs, led by Kagan, provided proof-of-principle for biological computing using neural cultures. Kagan demonstrated that *in vitro* neurons—including those derived from human iPSCs—could learn to play the video game Pong when interfaced with a simulated game environment through multi-electrode arrays[164]. This system, termed "DishBrain" proved that biological neurons exhibit goal-directed behavior and can improve their performance through real-time electrophysiological feedback, satisfying fundamental criteria for learning. Beyond advancing computational neuroscience, this finding confirms that biological neural networks can bidirectionally exchange information with digital systems, creating a critical proof-of-concept for hybrid intelligence.

Brainware and brain organoid reservoir computing

Cai and colleagues developed "Brainoware", a reservoir computing platform that harnesses the computational capacity of 3D biological neural networks[165]. In this system, brain organoids serve as computational reservoirs: they receive input signals through multi-electrode arrays and generate complex, high-dimensional output patterns that can be decoded for tasks such as speech recognition and nonlinear equation solving. Brainoware demonstrated that brain organoids can perform computations typically handled by artificial neural networks, while consuming far less energy and possessing inherent learning capabilities. Integrating the organoid intelligence concept with developmental neurotoxicity testing has expanded the application scope of brain organoid computing, as organoid intelligence approaches can assess how environmental toxins affect neural plasticity[142].

Brain–computer interfaces: bridging carbon and silicon

Brain–computer interfaces (BCIs) represent another parallel and convergent path toward integrating biological and artificial information processing. Technologies such as Neuralink have demonstrated that implantable electrode arrays enable high-bandwidth neural recording and stimulation, with emerging clinical applications in restoring motor function, communication, and sensory perception for patients with neurological injuries[166]. Willett and colleagues achieved a breakthrough in high-

performance neural prosthesis speech decoding, translating neural signals from the motor cortex into text at speeds approaching natural speech, validating the clinical feasibility of brain–computer communication[167]. Brain organoids provide a complementary platform for developing and testing BCIs within a human cellular context, allowing optimization of electrode–neuron interfaces and investigation of neural adaptations to chronic stimulation.

Patient-Avatar-Surrogate system: Toward the transparent human

The convergence of organoid intelligence, BCIs, digital twin technology, and traditional AI points toward a future encapsulated by the "Patient-Avatar" framework—a paradigm that integrates biological and digital representations of individual patients to advance personalized neuroscience and medicine. This framework comprises three interlinked components: (i) the biological component, consisting of patient-derived brain organoids that preserve the individual's genetic background, cellular responses, and drug sensitivities; (ii) the digital twin component, comprising computational models that simulate the patient's brain structure, connectivity, and disease trajectory using multimodal clinical data[168]; and (iii) the silicon-based AGI component, consisting of machine learning algorithms that integrate biological and digital data to predict treatment outcomes, optimize therapeutic strategies, and identify novel drug targets.

The "transparent human"—the vision in which the Patient-Avatar-Surrogate provides a comprehensive, multi-scale view of individual neurobiology from the molecular to the systems level—represents the ultimate goal of precision neurology. Patient-derived brain organoids provide biological ground truth, capturing unique cellular phenotypes and therapeutic vulnerabilities. Digital twin models informed by neuroimaging, genomics, and longitudinal clinical data provide the computational foundation for simulating disease progression and treatment response[169]. Silicon-based AGI systems integrate these data streams to generate actionable clinical insights unattainable from any single data source. This tripartite framework positions the Patient-Avatar not merely as a diagnostic or therapeutic tool, but as a new paradigm for understanding the relationship between individual human biology and computational intelligence.

Carbon–silicon synapses: Neuromorphic computing and mutual inspiration

Carbon-based biological intelligence and silicon-based AI do not interact unidirectionally; rather, they mutually inspire and enhance each other. Studies of biological neural computation—utilizing brain organoids, neural recordings, and computational neuroscience—continually inspire the design of neuromorphic computing architectures that emulate the efficiency and adaptability of biological synapses[170]. Neuromorphic chips, such as Intel's Loihi and IBM's TrueNorth, employ spiking neural network architectures that mimic the temporal dynamics and event-driven processing of biological neurons, achieving remarkable energy efficiency gains on specific computational tasks. Conversely, advances in AI—particularly large language models, reinforcement learning, and multimodal foundation models—provide powerful analytical tools for interpreting the complex datasets generated by brain organoid experiments, from single-cell transcriptomics to high-density electrophysiology.

Developing interfaces that enable seamless information transfer between biological and silicon substrates—termed "carbon–silicon synapses"—represents a grand challenge at the intersection of neuroscience, materials science, and computer engineering[14]. Envisioned hybrid intelligence systems would leverage the complementary strengths of each substrate: biological neural networks excel at energy-efficient learning, pattern recognition in noisy environments, and adaptive generalization, while silicon-based systems offer speed, precision, reproducibility, and scalability. Fusing these capabilities through the patient-avatar framework may fundamentally reshape not only precision medicine but also the broader landscape of intelligence research, blurring the boundaries between biological and artificial cognition.

Ethical considerations in the era of organoid intelligence

The rise of organoid intelligence and hybrid biocomputing systems raises profound ethical questions that the scientific community must proactively address. As brain organoids become increasingly complex, researchers must carefully evaluate whether they might develop features associated with consciousness, sentience, or moral status[171]. The Baltimore Declaration explicitly calls for the establishment of ethical frameworks governing organoid intelligence research, including guidelines on permissible organoid complexity, acceptable durations of neural activity recording, and the boundaries of human–machine neural interfaces. The International Society for Stem Cell Research has similarly emphasized the ongoing need for ethical oversight of brain organoid research, particularly as these systems acquire increasingly sophisticated computational and functional capabilities[172]. These ethical considerations—encompassing consciousness and sentience, international regulatory frameworks, newly issued Chinese national guidelines, cross-species chimeras, informed consent, and pathways toward harmonized global governance—will be comprehensively discussed in the next section.

Ethics, regulation, and policy in brain organoid research

The rapid maturation of brain organoid technology—from developmental models to systems capable of complex electrophysiological activity, circuit integration, and biological computing—urgently demands dedicated ethical frameworks and governance structures. While the ethical foundations of human stem cell research are firmly established through decades of international deliberation, brain organoids present unique challenges that extend beyond the traditional boundaries of stem cell ethics. These three-dimensional neural tissues raise distinctive questions concerning the potential emergence of consciousness-like states, the moral status of dynamically developing biological systems, the ethical boundaries of human–animal neural chimeras, and the adequacy of informed consent frameworks designed for static biological specimens rather than living, learning neural constructs[173,174]. This section provides a comprehensive review of the evolving ethical landscape, examines theoretical and empirical debates surrounding organoid consciousness, analyzes major international regulatory frameworks, including China's pioneering national guidelines, and proposes pathways toward harmonized global governance.

The evolving ethical landscape of brain organoids

Brain organoids occupy a unique ethical category that distinguishes them from both traditional two-dimensional cell

cultures and whole living organisms. Unlike conventional cell lines, brain organoids self-organize into three-dimensional structures that recapitulate aspects of human brain development, including the formation of progenitor zones, cortical lamination, and spontaneous network activity patterns resembling those observed in preterm infants[150, 171]. This structural and functional complexity has prompted a fundamental reassessment of the ethical categories traditionally used to govern neural tissue research. The shift from general stem cell ethics toward organoid-specific frameworks reflects the recognition that existing guidelines—designed primarily around embryonic stem cell derivation and cloning—are inadequate for addressing the distinctive attributes of organoid systems[173].

The dual-use nature of brain organoid research further amplifies these ethical complexities. On one hand, brain organoids serve as powerful platforms for disease modeling, drug discovery, and regenerative medicine, with clear therapeutic benefits. On the other hand, the emerging field of organoid intelligence positions these biological neural networks as substrates for biological computing, raising novel questions about the moral status of systems engaged in goal-directed computation and learning[175, 176]. The DishBrain and Brainoware platforms have already demonstrated that biological neural networks can perform computational tasks, including video game learning and reservoir computing, establishing proof-of-principle for integrating living neural tissue with digital systems[165, 177]. This convergence of therapeutic and computational applications creates an ethical landscape in which the same biological system can simultaneously function as a research tool, disease model, and computational substrate—while existing regulatory frameworks treat these categories as distinct.

The "Patient-Avatar" framework, integrating patient-derived brain organoids, digital twin models, and AI, further complicates this ethical domain by creating hybrid biological–digital representations of individual patients. These developments demand governance approaches capable of accommodating the dynamic, multifunctional nature of advanced brain organoids while upholding the ethical principles of beneficence, non-maleficence, autonomy, and justice that underpin biomedical research[49].

Consciousness, sentience, and moral status

The question of whether brain organoids might develop consciousness or consciousness-like states represents the most contentious ethical issue in the field. Current scientific consensus holds that no existing brain organoids possess consciousness comparable to an intact human brain[171]. Brain organoids lack the sensory input integration, bodily context, and subcortical structures considered necessary for full consciousness. However, the field acknowledges that organoid complexity is rapidly increasing, and current limitations provide no guarantee that this assessment is permanent[47]. Cortical organoids generate complex oscillatory patterns, including nested gamma oscillations that recapitulate electrophysiological features of the preterm human brain, while photosensitive organoids exhibit stimulus-dependent network responses[150, 151]. These findings, while not yet at the level of consciousness, indicate that the functional capacities of brain organoids are advancing in directions that require ongoing ethical deliberation.

Theoretical frameworks for understanding consciousness yield divergent predictions regarding the moral status of organoids. Integrated Information Theory (IIT) proposes that consciousness corresponds to a system's capacity to integrate information—a property quantified by the measure Φ (phi)—and suggests that any system with sufficient integrated information, regardless of substrate, possesses some degree of consciousness warranting moral consideration[174, 178]. Under IIT, brain organoids with complex recurrent connectivity and information integration capacity might be endowed with a rudimentary form of consciousness. In contrast, Global Workspace Theory (GWT) posits that consciousness requires a global workspace architecture capable of broadcasting selected information across distributed brain networks, which current organoids—lacking attentional mechanisms and the requisite large-scale structural organization—do not possess[165, 174]. These theoretical disagreements have direct governance implications: IIT would counsel a more precautionary approach to organoid research, while GWT would support greater permissiveness given current organoid limitations.

In response to these uncertainties, ethicists have proposed a precautionary principle framework that emphasizes ongoing monitoring rather than definitive categorical answers. Jeziorski et al. argue that the most defensible ethical stance is to acknowledge the "uncertain moral status" of advanced brain organoids—analogous to the moral status accorded to non-human animals or human fetuses—and to adopt anticipatory rather than speculative neuroethics[171]. Koplin and colleagues have proposed a three-tier moral framework: non-conscious organoids are regulated as human biological material; organoids with potential rudimentary consciousness receive protection from suffering; organoids with advanced cognitive capacities are monitored for emergent properties requiring additional safeguards[175]. Proposed benchmarks for consciousness-like states include self-monitoring capacity, behavioral flexibility, and affective responsiveness, with the Perturbational Complexity Index (PCI) emerging as a candidate measurement tool adapted from clinical consciousness assessment[178, 179]. In January 2025, an international team of neuroscientists, cognitive scientists, and philosophers published a manifesto in *Frontiers in Science* calling for the development of a universal consciousness test applicable across all substrates—biological, algorithmic, and hybrid—before the convergence of advanced AI and brain organoid technologies renders the question urgent[177].

International regulatory frameworks

The global regulatory landscape for human brain organoids, organoid chimeras, and embryo models reveals starkly divergent philosophical commitments and underlying ethical assumptions. No single jurisdiction has yet achieved comprehensive governance, and the patchwork of overlapping standards creates both opportunities for regulatory innovation and risks of ethical inconsistency. While the ISSCR provides influential international standards[173], national approaches vary considerably in their emphasis on precaution versus innovation. For a detailed comparative analysis of regulatory frameworks across the USA, EU, Australia, and China—including their divergent approaches to consciousness monitoring, chimera research, and ISEM oversight—see Ref. [180].

Overall, these frameworks converge on transparency, donor autonomy, and animal welfare but diverge on permissiveness toward chimera research, stringency of consciousness monitoring, and the balance between promoting innovation and preventing risk. This regulatory fragmentation poses particular challenges for

multinational research collaborations, where compliance with multiple jurisdictions may create conflicting ethical obligations.

China's pioneering organoid ethics governance

In April 2025, China issued the world's first comprehensive national-level guidelines dedicated to brain organoids, chimeras, and integrated stem cell-based embryo models (ISEMs)[181]. These guidelines introduce a distinctive governance model that integrates Confucian communitarian values with Western bioethical principles, establishing operational mechanisms such as dynamic consent protocols and domain-specific safeguards for high-risk research categories. For detailed analysis of the guidelines' three-tiered architecture, five foundational principles, and quantitative thresholds for consciousness monitoring, see Ref. [181].

As brain organoids continue to advance—toward more faithful recapitulation of human neural circuitry, longer maturation periods, and deeper integration with animal brains and computational systems—the window for establishing robust governance frameworks is narrowing. The convergence of brain organoid technology with AI and biological computing makes the development of governance that is anticipatory, adaptive, and internationally harmonized not merely an ethical aspiration but a practical necessity for responsible scientific progress.

The ethical governance of brain organoid research stands at a critical inflection point. Within a decade of their inception, these three-dimensional neural models have progressed from developmental curiosities to systems capable of complex electrophysiological activity, disease modeling, drug screening, and biological computing. This rapid functional advancement has outpaced the development of dedicated ethical frameworks, creating governance gaps that are particularly acute at the intersections of neuroscience, computer science, and international law.

The field has moved beyond the foundational stem cell ethics of the early 2000s toward organoid-specific frameworks that address consciousness potential, dual-use dilemmas, and hybrid biological–digital systems. Theoretical debates between IIT and GWT, precautionary tiered moral frameworks, and the emergence of candidate consciousness metrics such as PCI reflect growing sophistication in addressing the moral status of advanced neural tissue models.

Internationally, regulatory divergence remains pronounced, with the USA, EU, Australia, and China pursuing distinct governance philosophies. China's 2025 guidelines represent a pioneering experiment in centralized, anticipatory governance that directly confronts previously unregulated high-risk domains[180,181]. However, no jurisdiction has yet established comprehensive frameworks for organoid intelligence or biological computing, and the patchwork of national regulations creates compliance challenges for multinational research.

Future governance must balance innovation with ethical vigilance through anticipatory, adaptive, and internationally harmonized approaches. The window for establishing robust frameworks is narrowing as brain organoids advance toward more faithful recapitulation of human neural circuitry, longer maturation periods, and deeper integration with animal brains and computational systems. The convergence of biological and artificial intelligence—embodied in Patient-Avatar frameworks and biological computing platforms—demands governance that is as dynamic and multifunctional as the technologies it seeks to regulate. As the field prepares for the ISSCR's 2026 guidelines and anticipated regulatory updates across jurisdictions, active engagement of scientists, ethicists, policymakers, and public stakeholders will be essential to ensure that brain organoid research fulfills its therapeutic promise while respecting the profound ethical questions that these remarkable biological systems raise.

Conclusion and future perspectives

This review has charted the rapid progression of brain organoid research from fundamental developmental studies to clinical translation, drug development, biocomputing applications, and comprehensive ethical governance. Within a decade of their inception, these 3D models have become indispensable tools for studying both normal and pathological human brain function, opening research avenues that were previously inaccessible.

We have provided the first systematic overview in this review of normal brain organoid models, spanning developmental stages from fetal neurogenesis to postnatal-equivalent maturation, region-specific organoids of the hippocampus, thalamus, hypothalamus, and striatum, cerebellar organoids with functionally mature Purkinje cells, cerebrovascular and blood–brain barrier organoids, and chimeric cross-species integration models—collectively establishing the foundational reference architecture for all disease modeling. We have also comprehensively mapped the landscape of brain tumor organoid research, from glioblastoma and low-grade diffuse gliomas to meningiomas, vestibular schwannomas, pituitary tumors, and pediatric CNS tumors, including medulloblastoma and ependymoma, critically identifying both established models and persistent gaps. Our extended coverage of neurodegenerative, psychiatric, developmental, cerebrovascular, and traumatic disorders, together with a dedicated examination of ethical, regulatory, and policy frameworks, reflects the maturation of brain organoid technology across both biomedical and societal dimensions.

Several technical challenges persist. Most organoid models lack functional vasculature, limiting their size, maturation, and physiological relevance[172,173]. Despite progress in integrating microglia, oligodendrocytes, and vascular components, these elements remain underrepresented. Batch-to-batch variability continues to affect reproducibility despite improved protocols, and ethical considerations regarding potential consciousness or sentience in advanced organoids require ongoing deliberation.

The translational focus of the field has shifted decisively toward patient-derived materials—whether through iPSC reprogramming or direct surgical sample culture. This approach preserves individual genetic and disease characteristics, positioning brain organoids as pivotal tools for precision neurology.

Future advances are likely to arise from integrating brain organoids with single-cell multi-omics, spatial transcriptomics, optogenetics, and artificial intelligence. Standardized organoid atlases and international data-sharing initiatives can harmonize research efforts and accelerate clinical translation[182]. The Patient-Avatar framework—integrating biological organoids, digital twin models, and artificial general intelligence—envisions brain organoids not merely as experimental models but as computational interfaces bridging biological and machine intelligence[49,168].

In summary, brain organoid technology is reshaping neuroscience by providing experimentally accessible models of the

human brain. These systems now span the full spectrum of brain research—from normal developmental neurobiology to comprehensive disease modeling, clinical applications, biological computing, and ethical governance—and herald a future in which biological and artificial intelligence increasingly converge, where Patient-Avatar-Surrogate models may lead a new era of personalized brain medicine.

Research Ethics and Patient Consent

Not applicable.

Availability of data and materials

Not applicable.

Acknowledgements

During the preparation of this work, the authors used the K-Dense Web (k-dense.ai) for illustration support, and employed Zhipu GLM-5 and KIMI for English language polishing and literature verification. Gemini nano-banana 2 provided conceptual assistance for image design. All figures were ultimately designed and produced independently by the authors. The authors reviewed and edited all content as needed and take full responsibility for the content of this publication.

Funding

No funding for this research.

Declaration of conflicting interests

Financial support for this work was provided by Shanghai Lisheng Biotech Ltd (Lisheng). The authors declare that the manuscript was written with due regard for ethical and responsible conduct. X.X.H. is the founder of Lisheng. No other competing financial interests are declared by any of the authors.

Author contribution

Jianfei Long conducted a literature review on brain biology and performed manuscript editing. Xinxin Han designed the conceptual framework and performed manuscript writing. Both authors participated in the revision and final approval of this review.

References

[1] Hodge, R. D., Bakken, T. E., Miller, J. A., Smith, K. A., Barkan, E. R., Graybuck, L. T., Close, J. L., Long, B., Johansen, N., Penn, O. et al. Conserved cell types with divergent features in human versus mouse cortex. *Nature*, **2019**, 573(7772): 61–68. https://doi.org/10.1038/s41586-019-1506-7

[2] Pollen, A. A., Bhaduri, A., Andrews, M. G., Nowakowski, T. J., Meyerson, O. S., Mostajo-Radji, M. A., Di Lullo, E., Alvarado, B., Bedolli, M., Dougherty, M. L. et al. Establishing cerebral organoids as models of human-specific brain evolution. *Cell*, **2019**, 176(4): 743–756.e17. https://doi.org/10.1016/j.cell.2019.01.017

[3] Paşca, S. P. The rise of three-dimensional human brain cultures. *Nature*, **2018**, 553(7689): 437–445. https://doi.org/10.1038/nature25032

[4] Kanton, S., Boyle, M. J., He, Z. S., Santel, M., Weigert, A., Sanchís-Calleja, F., Guijarro, P., Sidow, L., Fleck, J. S., Han, D. D. et al. Organoid single-cell genomic atlas uncovers human-specific features of brain development. *Nature*, **2019**, 574(7778): 418–422. https://doi.org/10.1038/s41586-019-1654-9

[5] National Institutes of Health. NIH announces major policy shift to end use of human fetal tissue in NIH-supported research. NIH News Release. 2026. Availabel at https://www.nih.gov/news-events/news-releases/nih-announces-major-policy-shift-end-use-human-fetal-tissue-nih-supported-research (Access January 22, 2026)

[6] Lancaster, M. A., Renner, M., Martin, C. A., Wenzel, D., Bicknell, L. S., Hurles, M. E., Homfray, T., Penninger, J. M., Jackson, A. P., Knoblich, J. A. Cerebral organoids model human brain development and microcephaly. *Nature*, **2013**, 501(7467): 373–379. https://doi.org/10.1038/nature12517

[7] Eichmüller, O. L., Knoblich, J. A. Human cerebral organoids—a new tool for clinical neurology research. *Nature Reviews Neurology*, **2022**, 18(11): 661–680. https://doi.org/10.1038/s41582-022-00723-9

[8] Kelley, K. W., Paşca, S. P. Human brain organogenesis: Toward a cellular understanding of development and disease. *Cell*, **2022**, 185(1): 42–61. https://doi.org/10.1016/j.cell.2021.10.003

[9] Hubert, C. G., Read, R. D., Miller, T. E., Kim, U., Wang, X. et al. Modeling gliomas with organoids: Classification, fidelity, and guidelines for translational neuro-oncology. *Neuro-Oncology*, **2026**: noag085. https://doi.org/10.1093/neuonc/noag085

[10] Baisiwala, S., Fazzari, E., Li, M. X., Martija, A., Azizad, D. J., Sun, L., Herrera, G., Phan, T., Monteleone, A., Kan, R. L. et al. A human tumor-immune organoid model of glioblastoma. *Cell Reports*, **2026**, 45(1): 116790. https://doi.org/10.1016/j.celrep.2025.116790

[11] Jacob, F., Salinas, R. D., Zhang, D. Y., Nguyen, P. T. T., Schnoll, J. G., Wong, S. Z. H., Thokala, R., Sheikh, S., Saxena, D., Prokop, S. et al. A patient-derived glioblastoma organoid model and biobank recapitulates inter- and intra-tumoral heterogeneity. *Cell*, **2020**, 180(1): 188–204.e22. https://doi.org/10.1016/j.cell.2019.11.036

[12] Camp, J. G., Badsha, F., Florio, M., Kanton, S., Gerber, T., Wilsch-Bräuninger, M., Lewitus, E., Sykes, A., Hevers, W., Lancaster, M. et al. Human cerebral organoids recapitulate gene expression programs of fetal neocortex development. *Proceedings of the National Academy of Sciences of the United States of America*, **2015**, 112(51): 15672–15677. https://doi.org/10.1073/pnas.1520760112

[13] Lancaster, M. A., Knoblich, J. A. Generation of cerebral organoids from human pluripotent stem cells. *Nature Protocols*, **2014**, 9(10): 2329–2340. https://doi.org/10.1038/nprot.2014.158

[14] Smirnova, L., Hartung, T. The promise and potential of brain organoids. *Advanced Healthcare Materials*, **2024**, 13(21): 2302745. https://doi.org/10.1002/adhm.202302745

[15] Quadrato, G., Nguyen, T., Macosko, E. Z., Sherwood, J. L., Min Yang, S., Berger, D. R., Maria, N., Scholvin, J., Goldman, M., Kinney, J. P. et al. Cell diversity and network dynamics in photosensitive human brain organoids. *Nature*, **2017**, 545(7652): 48–53. https://doi.org/10.1038/nature22047

[16] Morales Pantoja, I. E., Smirnova, L., Muotri, A. R., Wahlin, K. J., Kahn, J., Boyd, J. L., Gracias, D. H., Harris, T. D., Cohen-Karni, T., Caffo, B. S. et al. First Organoid Intelligence (OI) workshop to form an OI community. *Frontiers in Artificial Intelligence*, **2023**, 6: 1116870. https://doi.org/10.3389/frai.2023.1116870

[17] Gordon, A., Yoon, S. J., Tran, S. S., Makinson, C. D., Park, J. Y., Andersen, J., Valencia, A. M., Horvath, S., Xiao, X. S., Huguenard, J. R. et al. Long-term maturation of human cortical organoids matches key early postnatal transitions. *Nature Neuroscience*, **2021**, 24(3): 331–342. https://doi.org/10.1038/s41593-021-00802-y

[18] Faravelli, I., Antón-Bolaños, N., Wei, A., Faits, T., Sampath Kumar, A., Andreadis, S., Kastli, R., Montero Crespo, M., Steiger, M., Leible, D., et al. Human brain organoids record the passage of

time over multiple years in culture. bioRxiv, **2025**: 10.01.679721. https://doi.org/10.1101/2025.10.01.679721

[19] Qian, X. Y., Su, Y. J., Adam, C. D., Deutschmann, A. U., Pather, S. R., Goldberg, E. M., Su, K. N., Li, S. Y., Lu, L., Jacob, F. et al. Sliced human cortical organoids for modeling distinct cortical layer formation. *Cell Stem Cell*, **2020**, 26(5): 766–781.e9. https://doi.org/10.1016/j.stem.2020.02.002

[20] Giandomenico, S. L., Mierau, S. B., Gibbons, G. M., Wenger, L. M. D., Masullo, L., Sit, T., Sutcliffe, M., Boulanger, J., Tripodi, M., Derivery, E. et al. Cerebral organoids at the air–liquid interface generate diverse nerve tracts with functional output. *Nature Neuroscience*, **2019**, 22(4): 669–679. https://doi.org/10.1038/s41593-019-0350-2

[21] Petersilie, L., Heiduschka, S., Nelson, J. S. E., Neu, L. A., Le, S., Anand, R., Kafitz, K. W., Prigione, A., Rose, C. R. Cortical brain organoid slices (cBOS) for the study of human neural cells in minimal networks. *iScience*, **2024**, 27(4): 109415. https://doi.org/10.1016/j.isci.2024.109415

[22] He, Z. S., Dony, L., Fleck, J. S., Szałata, A., Li, K. X., Slišković, I., Lin, H. C., Santel, M., Atamian, A., Quadrato, G. et al. An integrated transcriptomic cell atlas of human neural organoids. *Nature*, **2024**, 635(8039): 690–698. https://doi.org/10.1038/s41586-024-08172-8

[23] Hendriks, D., Pagliaro, A., Andreatta, F., Ma, Z. L., van Giessen, J., Massalini, S., López-Iglesias, C., van Son, G. J. F., DeMartino, J., Damen, J. M. A. et al. Human fetal brain self-organizes into long-term expanding organoids. *Cell*, **2024**, 187(3): 712–732.e38. https://doi.org/10.1016/j.cell.2023.12.012

[24] Smits, L. M., Reinhardt, L., Reinhardt, P., Glatza, M., Monzel, A. S., Stanslowsky, N., Rosato-Siri, M. D., Zanon, A., Antony, P. M., Bellmann, J., et al. Modeling Parkinson's disease in midbrain-like organoids. *NPJ Parkinsons Disease*, **2019**, 5(1): 5. https://doi.org/10.1038/s41531-019-0078-4

[25] Birtele, M., Storm, P., Sharma, Y., Kajtez, J., Wahlestedt, J. N., Sozzi, E., Nilsson, F., Stott, S., He, X. L., Mattsson, B. et al. Single-cell transcriptional and functional analysis of dopaminergic neurons in organoid-like cultures derived from human fetal midbrain. *Development*, **2022**, 149(23): dev200504. https://doi.org/10.1242/dev.200504

[26] Sakaguchi, H., Kadoshima, T., Soen, M., Narii, N., Ishida, Y., Ohgushi, M., Takahashi, J., Eiraku, M., Sasai, Y. Generation of functional hippocampal neurons from self-organizing human embryonic stem cell-derived dorsomedial telencephalic tissue. *Nature Communications*, **2015**, 6: 8896. https://doi.org/10.1038/ncomms9896

[27] Wu, Y., Cheng, J. H., Qi, J., Hang, C., Dong, R. H., Low, B. C., Yu, H., Jiang, X. Y. Three-dimensional liquid metal-based neuro-interfaces for human hippocampal organoids. *Nature Communications*, **2024**, 15: 4047. https://doi.org/10.1038/s41467-024-48452-5

[28] Xiang, Y. F., Tanaka, Y., Cakir, B., Patterson, B., Kim, K. Y., Sun, P. N., Kang, Y. J., Zhong, M., Liu, X. R., Patra, P. et al. hESC-derived thalamic organoids form reciprocal projections when fused with cortical organoids. *Cell Stem Cell*, **2019**, 24(3): 487–497.e7. https://doi.org/10.1016/j.stem.2018.12.015

[29] Duenki, T., Ikeuchi, Y. Multi-organoid loop cerebral connectoids exhibit enhanced neuronal network dynamics and sequence-specific entrainment. *Communications Biology*, **2026**, 9: 302. https://doi.org/10.1038/s42003-026-09589-9

[30] Patton, M. H., Thomas, K. T., Bayazitov, I. T., Newman, K. D., Kurtz, N. B., Robinson, C. G., Ramirez, C. A., Trevisan, A. J., Bikoff, J. B., Peters, S. T. et al. Synaptic plasticity in human thalamocortical assembloids. *Cell Reports*, 43(8): 114503. https://doi.org/10.1016/j.celrep.2024.114503

[31] Huang, W. K., Wong, S. Z. H., Pather, S. R., Nguyen, P. T. T., Zhang, F., Zhang, D. Y., Zhang, Z. J., Lu, L., Fang, W. Q., Chen, L. Y. et al. Generation of hypothalamic arcuate organoids from human induced pluripotent stem cells. *Cell Stem Cell*, **2021**, 28(9): 1657–1670.e10. https://doi.org/10.1016/j.stem.2021.04.006

[32] Wang, S. J., Jiang, D. Y., Xiao, Y., Qin, Q. Z., Zhang, H. Y., Ye, L. T., Jin, J. D., Jiang, X. X., Guo, Q. H. Human pituitary organoids: Transcriptional landscape deciphered by scRNA-seq and stereo-seq, with insights into SOX3's role in pituitary development. *Advanced Science*, **2025**, 12(14): 2414230. https://doi.org/10.1002/advs.202414230

[33] Miura, Y., Li, M. Y., Birey, F., Ikeda, K., Revah, O., Thete, M. V., Park, J. Y., Puno, A., Lee, S. H., Porteus, M. H. et al. Generation of human striatal organoids and cortico-striatal assembloids from human pluripotent stem cells. *Nature Biotechnology*, **2020**, 38(12): 1421–1430. https://doi.org/10.1038/s41587-020-00763-w

[34] Pellegrini, L., Bonfio, C., Chadwick, J., Begum, F., Skehel, M., Lancaster, M. A. Human CNS barrier-forming organoids with cerebrospinal fluid production. *Science*, 369(6500): eaaz5626. https://doi.org/10.1126/science.aaz5626

[35] Budinger, D., Puigdevall, P., Hall, G. T., Roth, C., Xenakis, T., Marrosu, E., Jerber, J., Di Domenico, A., Picco, F., Kilpinen, H. et al. An *in vivo* and *in vitro* spatiotemporal profile of human midbrain development. *Nature Communications*, **2026**, 17: 1354. https://doi.org/10.1038/s41467-025-67779-1

[36] Wang, X. Y., Sun, G. Y., Tang, M. M., Li, D., Qi, J. H., Wang, C. Y., Wang, Y. K., Hu, B. Y. Human midbrain organoids enriched with dopaminergic neurons for long-term functional evaluation. *Cell Proliferation*, **2025**, 58(7): e70005. https://doi.org/10.1111/cpr.70005

[37] Muguruma, K., Nishiyama, A., Kawakami, H., Hashimoto, K., Sasai, Y. Self-organization of polarized cerebellar tissue in 3D culture of human pluripotent stem cells. *Cell Reports*, **2015**, 10(4): 537–550. https://doi.org/10.1016/j.celrep.2014.12.051

[38] Atamian, A., Birtele, M., Hosseini, N., Nguyen, T., Seth, A., Del Dosso, A., Paul, S., Tedeschi, N., Taylor, R., Coba, M. P. et al. Human cerebellar organoids with functional Purkinje cells. *Cell Stem Cell*, **2024**, 31(1): 39–51.e6. https://doi.org/10.1016/j.stem.2023.11.013

[39] Apsley, E. J., Riepsaame, J., Cheng, Y. C., Cowley, S. A., Becker, E. B. E. Cerebellar organoids model cell type-specific*FOXP2*expression during human cerebellar development. *Disease Models & Mechanisms*, **2025**, 18(11): dmm052290. https://doi.org/10.1242/dmm.052290

[40] Chen, Y., Bury, L. A., Chen, F., Aldinger, K. A., Miranda, H. C., Wynshaw-Boris, A. Generation of advanced cerebellar organoids for neurogenesis and neuronal network development. *Human Molecular Genetics*, **2023**, 32(18): 2832–2841. https://doi.org/10.1093/hmg/ddad110

[41] Silva, T. P., Sousa-Luís, R., Fernandes, T. G., Bekman, E. P., Rodrigues, C. A. V., Vaz, S. H., Moreira, L. M., Hashimura, Y., Jung, S., Lee, B. et al. Transcriptome profiling of human pluripotent stem cell-derived cerebellar organoids reveals faster commitment under dynamic conditions. *Biotechnology and Bioengineering*, **2021**, 118(7): 2781–2803. https://doi.org/10.1002/bit.27797

[42] Nayler, S., Agarwal, D., Curion, F., Bowden, R., Becker, E. B. E. High-resolution transcriptional landscape of xeno-free human induced pluripotent stem cell-derived cerebellar organoids. *Scientific Reports*, **2021**, 11: 12959. https://doi.org/10.1038/s41598-021-91846-4

[43] Jalilian, E., Shin, S. R. Novel model of cortical–meningeal organoid co-culture system improves human cortical brain organoid cytoarchitecture. *Scientific Reports*, **2023**, 13: 7809. https://doi.org/10.1038/s41598-023-35077-9

[44] Mansour, A. A., Gonçalves, J. T., Bloyd, C. W., Li, H., Fernandes, S., Quang, D., Johnston, S., Parylak, S. L., Jin, X., Gage, F. H. An *in vivo* model of functional and vascularized human brain organoids. *Nature Biotechnology*, **2018**, 36(5): 432–441. https://doi.org/10.1038/nbt.4127

[45] Shi, Y. C., Sun, L., Wang, M. D., Liu, J. W., Zhong, S. J., Li, R., Li, P., Guo, L. J., Fang, A., Chen, R. G. et al. Vascularized human cortical organoids (vOrganoids) model cortical development *in vivo*. *PLoS Biology*, **2020**, 18(5): e3000705. https://doi.org/10.1371/journal.pbio.3000705

[46] Cakir, B., Xiang, Y. F., Tanaka, Y., Kural, M. H., Parent, M., Kang, Y. J., Chapeton, K., Patterson, B., Yuan, Y. F., He, C. S. et al. Engineering of human brain organoids with a functional vascular-like system. *Nature Methods*, **2019**, 16(11): 1169–1175. https://doi.org/10.1038/s41592-019-0586-5

[47] Revah, O., Gore, F., Kelley, K. W., Andersen, J., Sakai, N., Chen, X. Y., Li, M. Y., Birey, F., Yang, X., Saw, N. L. et al. Maturation and circuit integration of transplanted human cortical organoids. *Nature*, **2022**, 610(7931): 319–326. https://doi.org/10.1038/s41586-022-05277-w

[48] Stanton, A. E., Bubnys, A., Agbas, E., James, B., Park, D. S., Jiang, A. L., et al. Engineered 3D immuno-glial-neurovascular human miBrain model. *Proceedings of the National Academy of Sciences of the United States of America*, **2025**, 122(42): e2511596122. https://doi.org/10.1073/pnas.2511596122

[49] Pavlou, G., Spitz, S., Pramotton, F. M., Tsai, A., Li, B. M., Wang, X., Barr, O. M., Ko, E. C., Zhang, S., Ashley, S. J. et al. Engineered 3D human neurovascular model of Alzheimer's disease to study vascular dysfunction. *Biomaterials*, **2025**, 314: 122864. https://doi.org/10.1016/j.biomaterials.2024.122864

[50] Jgamadze, D., Lim, J. T., Zhang, Z. J., Harary, P. M., Germi, J., Mensah-Brown, K., Adam, C. D., Mirzakhalili, E., Singh, S., Ben Gu, J. et al. Structural and functional integration of human forebrain organoids with the injured adult rat visual system. *Cell Stem Cell*, **2023**, 30(2): 137–152.e7. https://doi.org/10.1016/j.stem.2023.01.004

[51] Dong, X., Xu, S. B., Chen, X., Tao, M. D., Tang, X. Y., Fang, K. H., Xu, M., Pan, Y. F., Chen, Y. J., He, S. J. et al. Human cerebral organoids establish subcortical projections in the mouse brain after transplantation. *Molecular Psychiatry*, **2021**, 26(7): 2964–2976. https://doi.org/10.1038/s41380-020-00910-4

[52] Kitahara, T., Sakaguchi, H., Morizane, A., Kikuchi, T., Miyamoto, S., Takahashi, J. Axonal extensions along corticospinal tracts from transplanted human cerebral organoids. *Stem Cell Reports*, **2020**, 15(2): 467–481. https://doi.org/10.1016/j.stemcr.2020.06.016

[53] Knight, G. T., Lundin, B. F., Iyer, N., Ashton, L. M., Sethares, W. A., Willett, R. M., Ashton, R. S. Engineering induction of singular neural rosette emergence within hPSC-derived tissues. *eLife*, **2018**, 7: e37549. https://doi.org/10.7554/elife.37549

[54] Li, C., Fleck, J. S., Martins-Costa, C., Burkard, T. R., Themann, J., Stuempflen, M., Peer, A. M., Vertesy, Á., Littleboy, J. B., Esk, C. et al. Single-cell brain organoid screening identifies developmental defects in autism. *Nature*, **2023**, 621(7978): 373–380. https://doi.org/10.1038/s41586-023-06473-y

[55] Meng, X. L., Reis, N., Bassik, M. C., Paşca, S. P. CRISPR screens in human neural organoids and assembloids. *Nature Protocols*, **2025**. https://doi.org/10.1038/s41596-025-01299-6

[56] Huang, R. E., Anand, G. M., Megale, H. C., Chen, J., Abraham-Igwe, C., Ramanathan, S. Arrayed single-gene perturbations identify drivers of human anterior neural tube closure. *eLife*, **2025**, 14: RP108224. https://doi.org/10.7554/eLife.108224.2

[57] She, R., Fair, T., Schaefer, N. K., Saunders, R. A., Pavlovic, B. J., Weissman, J. S., Pollen, A. A. Comparative landscape of genetic dependencies in human and chimpanzee stem cells. *Cell*, **2023**, 186(14): 2977–2994.e23. https://doi.org/10.1016/j.cell.2023.05.043

[58] Vassalou, S., Lalioti, M. E., Di Giaimo, R., Taraviras, S., Cappello, S., Kyrousi, C. Modelling human adult V-SVZ niche assembly and ependymal cell generation in brain organoids. *EMBO Reports*, **2026**, 27(1): 31–49. https://doi.org/10.1038/s44319-025-00621-3

[59] Logun, M., Wang, X., Sun, Y. S., Bagley, S. J., Li, N. N., Desai, A., Zhang, D. Y., Nasrallah, M. P., Pai, E. L., Oner, B. S. et al. Patient-derived glioblastoma organoids as real-time avatars for assessing responses to clinical CAR-T cell therapy. *Cell Stem Cell*, **2025**, 32(2): 181–190.e4. https://doi.org/10.1016/j.stem.2024.11.010

[60] Goranci-Buzhala, G., Mariappan, A., Gabriel, E., Ramani, A., Ricci-Vitiani, L., Buccarelli, M., D'Alessandris, Q. G., Pallini, R., Gopalakrishnan, J. Rapid and efficient invasion assay of glioblastoma in human brain organoids. *Cell Reports*, **2020**, 31(10): 107738. https://doi.org/10.1016/j.celrep.2020.107738

[61] Krieger, T. G., Tirier, S. M., Park, J., Jechow, K., Eisemann, T., Peterziel, H., Angel, P., Eils, R., Conrad, C. Modeling glioblastoma invasion using human brain organoids and single-cell transcriptomics. *Neuro-Oncology*, **2020**, 22(8): 1138–1149. https://doi.org/10.1093/neuonc/noaa091

[62] Bian, S., Repic, M., Guo, Z. M., Kavirayani, A., Burkard, T., Bagley, J. A., Krauditsch, C., Knoblich, J. A. Genetically engineered cerebral organoids model brain tumor formation. *Nature Methods*, **2018**, 15(8): 631–639. https://doi.org/10.1038/s41592-018-0070-7

[63] Ge, W. H., Kan, R. L., Yilgor, C., Fazzari, E., Nano, P. R., Azizad, D. J., Shinglot, H., Li, M., Ito, J. Y., Tse, C. et al. Human organoid tumor transplantation identifies functional glioblastoma-microenvironment communication mediated by PTPRZ1. *Cell Reports*, **2026**, 45(1): 116848. https://doi.org/10.1016/j.celrep.2025.116848

[64] Zheng, C. J., Wang, P., Zhang, D. L., Fang, Z., Feng, Y. T., Chen, J., Chen, J. H., Fu, Y. W., Yang, B., Yu, S. Q. et al. A novel organoid model retaining the glioma microenvironment for personalized drug screening and therapeutic evaluation. *Bioactive Materials*, **2025**, 53: 205–217. https://doi.org/10.1016/j.bioactmat.2025.07.015

[65] Park, J., Kim, D., Park, H. C., Park, M., Zhang, S. Z., Na, O., Lee, Y. S., Lee, M., Ahn, S., Chung, Y. J. Tumor microenvironment-preserving gliosarcoma organoids as an *in vitro* preclinical platform: A comparative analysis with glioblastoma models. *Journal of Translational Medicine*, **2025**, 23: 915. https://doi.org/10.1186/s12967-025-06952-y

[66] Hubert, C. G., Rivera, M., Spangler, L. C., Wu, Q. L., Mack, S. C., Prager, B. C., Couce, M., McLendon, R. E., Sloan, A. E., Rich, J. N. A three-dimensional organoid culture system derived from human glioblastomas recapitulates the hypoxic gradients and cancer stem cell heterogeneity of tumors FoundIn vivo. *Cancer Research*, **2016**, 76(8): 2465–2477. https://doi.org/10.1158/0008-5472.can-15-2402

[67] Kim, O., Sergi, Z., Yu, G. Y., Yamamoto, K., Quezado, M., Abdullaev, Z., Crooks, D. R., Kishimoto, S., Li, Q., Lu, P. et al. A patient-derived cell model for malignant transformation in IDH-mutant glioma. *Acta Neuropathologica Communications*, **2024**, 12: 148. https://doi.org/10.1186/s40478-024-01860-6

[68] Raleigh, D., Mirchia, K., Oten, S., Picart, T., Nguyen, M., Ambati, V., Vasudevan, H., Young, J., Taylor, J., Krishna, S. et al. Spatial synaptic connectivity underlies oligodendroglioma evolution and recurrence. *Research Square*, **2025**.

[69] Braun, F. K., Rothhammer-Hampl, T., Lorenz, J., Pohl, S., Menevse, A. N., Vollmann-Zwerenz, A., Bumes, E., Büttner, M., Zoubaa, S., Proescholdt, M. et al. Scaffold-based (matrigel™) 3D culture technique of glioblastoma recovers a patient-like immunosuppressive phenotype. *Cells*, **2023**, 12(14): 1856. https://doi.org/10.3390/cells12141856

[70] Bessler, N., Wezenaar, A. K. L., Ariese, H. C. R., Honhoff, C., Dommann, N., Wehrens, E. J., Ruiz Moreno, C., van den Broek, T. J. M., Collot, R. V. U., Kloosterman, D. J. et al. *De novo* H3.3K27M-altered diffuse midline glioma in human brainstem organoids to dissect GD2 CAR T cell function. *Nature Cancer*, **2026**, 7(2): 316–333. https://doi.org/10.1038/s43018-025-01084-0

[71] Larson, J. D., Kasper, L. H., Paugh, B. S., Jin, H. J., Wu, G., Kwon, C. H., Fan, Y. P., Shaw, T. I., Silveira, A. B., Qu, C. X. et al. Histone H3.*3 K27M accelerates spontaneous brainstem glioma and*

drives restricted changes in bivalent gene expression. Cancer Cell, **2019**, 35(1): 140–155.e7. https://doi.org/10.1016/j.ccell.2018.11.015

[72] Bressan, R. B., Southgate, B., Ferguson, K. M., Blin, C., Grant, V., Alfazema, N., Wills, J. C., Marques-Torrejon, M. A., Morrison, G. M., Ashmore, J. et al. Regional identity of human neural stem cells determines oncogenic responses to histone H3.*3 mutants. Cell Stem Cell*, **2021**, 28(5): 877–893.e9. https://doi.org/10.1016/j.stem.2021.01.016

[73] Zohdy, Y. M., Jahangiri, A., Jacob, F., Aksionau, A., Alawieh, A. M., Tong, A., Chern, B., Maldonado, J., Hoang, K., Nduom, E. et al. Patient-derived meningioma organoids: A reliable model for studying human tumor pathophysiology. *Cancers*, **2025**, 17(3): 526. https://doi.org/10.3390/cancers17030526

[74] Kim, D., Park, J., Park, H. C., Zhang, S. Z., Park, M., Park, S. A., Lee, S. H., Lee, Y. S., Park, J. S., Jeun, S. S. et al. Establishment of tumor microenvironment-preserving organoid model from patients with intracranial meningioma. *Cancer Cell International*, **2024**, 24: 36. https://doi.org/10.1186/s12935-024-03225-4

[75] Catasús, N., Casals-Sendra, G., Torres-Martin, M., Rosas, I., Kuebler, B., Mazuelas, H., Amilibia, E., Aran, B., Veiga, A., Raya, Á. et al. iPSC-based merlin-deficient Schwann cell-like spheroids as an *in vitro* system for studying NF2 pathogenesis. *Genes & Diseases*, **2025**, 12(6): 101615. https://doi.org/10.1016/j.gendis.2025.101615

[76] Chakrabarti, J., Pandey, R., Churko, J. M., Eschbacher, J., Mallick, S., Chen, Y. L., Hermes, B., Mallick, P., Stansfield, B. N., Pond, K. W. et al. Development of human pituitary neuroendocrine tumor organoids to facilitate effective targeted treatments of Cushing's disease. *Cells*, **2022**, 11(21): 3344. https://doi.org/10.3390/cells11213344

[77] Zhang, D. Y., Hugo, W., Redublo, P., Miao, H., Bergsneider, M., Wang, M. B., Kim, W., Yong, W. H., Heaney, A. P. A human ACTH-secreting corticotroph tumoroid model Novel Human ACTH-Secreting Tumor Cell *in vitro* Model. *eBioMedicine*, **2021**, 66: 103294. https://doi.org/10.1016/j.ebiom.2021.103294

[78] Mallick, S., Chakrabarti, J., Eschbacher, J., Moraitis, A. G., Greenstein, A. E., Churko, J., Pond, K. W., Livolsi, A., Thorne, C. A., Little, A. S. et al. Genetically engineered human pituitary corticotroph tumor organoids exhibit divergent responses to glucocorticoid receptor modulators. *Translational Research*, **2023**, 256: 56–72. https://doi.org/10.1016/j.trsl.2023.01.002

[79] Chen, D. T., Lei, T., Wang, Y. L., Yu, Z. T., Liu, S. Q., Ye, L., Li, W. F., Yang, Q., Jin, H. T., Liu, F. J. et al. Isolation and characterization of the adamantinomatous craniopharyngioma primary cells with cancer-associated fibroblast features. *Biomedicines*, **2025**, 13(4): 912. https://doi.org/10.3390/biomedicines13040912

[80] Apps, J. R., Martinez-Barbera, J. P. Genetically engineered mouse models of craniopharyngioma: An opportunity for therapy development and understanding of tumor biology. *Brain Pathology*, **2017**, 27(3): 364–369. https://doi.org/10.1111/bpa.12501

[81] Ballabio, C., Anderle, M., Gianesello, M., Lago, C., Miele, E., Cardano, M., Aiello, G., Piazza, S., Caron, D., Gianno, F. et al. Modeling medulloblastoma *in vivo* and with human cerebellar organoids. *Nature Communications*, **2020**, 11: 583. https://doi.org/10.1038/s41467-019-13989-3

[82] Ballabio, C., Gianesello, M., Lago, C., Okonechnikov, K., Anderle, M., Aiello, G., Antonica, F., Zhang, T. T., Gianno, F., Giangaspero, F. et al. Notch1 switches progenitor competence in inducing medulloblastoma. *Science Advances*, **2021**, 7(26): eabd2781. https://doi.org/10.1126/sciadv.abd2781

[83] Lago, C., Gianesello, M., Santomaso, L., Leva, G., Ballabio, C., Anderle, M., Antonica, F., Tiberi, L. Medulloblastoma and high-grade glioma organoids for drug screening, lineage tracing, co-culture and *in vivo* assay. *Nature Protocols*, **2023**, 18(7): 2143–2180. https://doi.org/10.1038/s41596-023-00839-2

[84] Lilly, J. V., Rokita, J. L., Mason, J. L., Patton, T., Stefankiewiz, S., Higgins, D., Trooskin, G., Larouci, C. A., Arya, K., Appert, E. et al. The children's brain tumor network (CBTN) - Accelerating research in pediatric central nervous system tumors through collaboration and open science. *Neoplasia*, **2023**, 35: 100846. https://doi.org/10.1016/j.neo.2022.100846

[85] Paassen, I., Williams, J., Ríos Arceo, C., Ringnalda, F., Mercer, K. S., Buhl, J. L., Moreno, N., Federico, A., Franke, N. E., Kranendonk, M. et al. Atypical teratoid/rhabdoid tumoroids reveal subgroup-specific drug vulnerabilities. *Oncogene*, **2023**, 42(20): 1661–1671. https://doi.org/10.1038/s41388-023-02681-y

[86] Li, S. J., Ren, J., Wu, J. N., Xia, Z. G., Li, Y. Z., Li, C. X., Cao, W. J. Establishment and molecular characterisation of patient-derived organoids for primary central nervous system lymphoma. *Leukemia*, **2025**, 39(5): 1169–1183. https://doi.org/10.1038/s41375-025-02562-1

[87] Gonzalez, C., Armijo, E., Bravo-Alegria, J., Becerra-Calixto, A., Mays, C. E., Soto, C. Modeling amyloid beta and tau pathology in human cerebral organoids. *Molecular Psychiatry*, **2018**, 23(12): 2363–2374. https://doi.org/10.1038/s41380-018-0229-8

[88] Blanchard, J. W., Akay, L. A., Davila-Velderrain, J., von Maydell, D., Mathys, H., Davidson, S. M., Effenberger, A., Chen, C. Y., Maner-Smith, K., Hajjar, I. et al. APOE4 impairs myelination *via* cholesterol dysregulation in oligodendrocytes. *Nature*, **2022**, 611(7937): 769–779. https://doi.org/10.1038/s41586-022-05439-w

[89] Zivko, C., Sagar, R., Xydia, A., Lopez-Montes, A., Mintzer, J., Rosenberg, P. B., Shade, D. M., Porsteinsson, A. P., Lyketsos, C. G., Mahairaki, V. iPSC-derived hindbrain organoids to evaluate escitalopram oxalate treatment responses targeting neuropsychiatric symptoms in Alzheimer's disease. *Molecular Psychiatry*, **2024**, 29(11): 3644–3652. https://doi.org/10.1038/s41380-024-02629-y

[90] Qiao, H. W., Zhao, W., Guo, M. J., Zhu, L. L., Chen, T., Wang, J. B., Xu, X. D., Zhang, Z. T., Wu, Y., Chen, P. Cerebral organoids for modeling of HSV-1-induced-amyloid β associated neuropathology and phenotypic rescue. *International Journal of Molecular Sciences*, **2022**, 23(11): 5981. https://doi.org/10.3390/ijms23115981

[91] Chen, X. W., Sun, G. Q., Tian, E., Zhang, M. Z., Davtyan, H., Beach, T. G., Reiman, E. M., Blurton-Jones, M., Holtzman, D. M., Shi, Y. H. Modeling sporadic Alzheimer's disease in human brain organoids under serum exposure. *Advanced Science*, **2021**, 8(18): 2101462. https://doi.org/10.1002/advs.202101462

[92] Nguyen-Vi, M., Julien, S., Janani, R., Meghna, M., Paula, L., Eric, D., Gilles, M., Michael, N., Chen Carol X-, Q., Narges, A. et al. Midbrain organoids with an SNCA gene triplication model key features of synucleinopathy. *Brain Communications*, 3(4): fcab223. https://doi.org/10.1093/braincomms/fcab223

[93] Jo, J., Xiao, Y. X., Sun, A. X., Cukuroglu, E., Tran, H. D., Göke, J., Tan, Z. Y., Saw, T. Y., Tan, C. P., Lokman, H. et al. Midbrain-like organoids from human pluripotent stem cells contain functional dopaminergic and neuromelanin-producing neurons. *Cell Stem Cell*, **2016**, 19(2): 248–257. https://doi.org/10.1016/j.stem.2016.07.005

[94] Gerasimova, E., Beenen, A. C., Kachkin, D., Regensburger, M., Zundler, S., Blumenthal, D. B., Lutzny-Geier, G., Winner, B., Prots, I. Novel co-culture model of T cells and midbrain organoids for investigating neurodegeneration in Parkinson's disease. *npj Parkinson's Disease*, **2025**, 11: 36. https://doi.org/10.1038/s41531-025-00882-8

[95] Kurniawan, H., Nickels, S. L., Zagare, A., Zuccoli, E., Rosety, I., Gomez-Giro, G., Glaab, E., Schwamborn, J. C. The Parkinson's disease-associated LRRK2-G2019S variant restricts serine metabolism, leading to microglial inflammation and dopaminergic neuron degeneration. *Journal of Neuroinflammation*, **2025**, 22: 244. https://doi.org/10.1186/s12974-025-03577-2

[96] Zagare, A., Balaur, I., Rougny, A., Saraiva, C., Gobin, M., Monzel, A. S., Ghosh, S., Satagopam, V. P., Schwamborn, J. C.

Deciphering shared molecular dysregulation across Parkinson's disease variants using a multi-modal network-based data integration and analysis. *npj Parkinson's Disease*, **2025**, 11: 63. https://doi.org/10.1038/s41531-025-00914-3

[97] Nishijima, A., Yokota, M., Kakuta, S., Yamaguchi, A., Ishikawa, K. I., Okano, H., Akamatsu, W., Hattori, N., Koike, M. Human midbrain organoids reveal the characteristics of axonal mitochondria specific to dopaminergic neurons. *Molecular Brain*, **2026**, 19: 8. https://doi.org/10.1186/s13041-025-01268-w

[98] Trapecar, M., Wogram, E., Svoboda, D., Communal, C., Omer, A., Lungjangwa, T., Sphabmixay, P., Velazquez, J., Schneider, K., Wright, C. W. et al. Human physiomimetic model integrating microphysiological systems of the gut, liver, and brain for studies of neurodegenerative diseases. *Science Advances*, **2021**, 7(5): eabd1707. https://doi.org/10.1126/sciadv.abd1707

[99] Vadodaria, K. C., Ji, Y., Skime, M., Paquola, A., Nelson, T., Hall-Flavin, D., Fredlender, C., Heard, K. J., Deng, Y. L., Le, A. T. et al. Serotonin-induced hyperactivity in SSRI-resistant major depressive disorder patient-derived neurons. *Molecular Psychiatry*, **2019**, 24(6): 795–807. https://doi.org/10.1038/s41380-019-0363-y

[100] Li, M. J., Duan, W., Hao, X. Y., Li, S. J., Wang, C., Liang, Y. Y., Hu, Z. W., Ma, D. R., Guo, M. N., Zuo, C. Y. et al. Effects of esketamine on electrophysiology and metabolic reprogramming in brain organoids: Insights into antidepressant mechanisms. *Molecular Psychiatry*, **2025**, 30(12): 6107–6118. https://doi.org/10.1038/s41380-025-03198-4

[101] Lu, K. Q., Hong, Y., Tao, M. D., Shen, L. P., Zheng, Z. L., Fang, K. H., Yuan, F., Xu, M., Wang, C., Zhu, D. Y. et al. Depressive patient-derived GABA interneurons reveal abnormal neural activity associated with HTR2C. *EMBO Molecular Medicine*, **2023**, 15: EMMM202216364. https://doi.org/10.15252/emmm.202216364

[102] El Soufi El Sabbagh, D., Kolinski Machado, A., Pappis, L., Leigh Beroncal, E., Ji, D., Nader, G., Ravi Chander, P., Choi, J., Duong, A., Jeong, H. et al. iPSC-derived cerebral organoids reveal mitochondrial, inflammatory and neuronal vulnerabilities in bipolar disorder. *Translational Psychiatry*, **2025**, 15: 315. https://doi.org/10.1038/s41398-025-03529-7

[103] Cheng, K., Williams, A., Kshirsagar, A., Kulkarni, S., Karmacharya, R., Massachusetts, M. H. B., Kim, D. H. et al. Machine learning-enabled detection of electrophysiological signatures in iPSC-derived models of schizophrenia and bipolar disorder. *APL Bioengineering*, **2025**, 9(3): 036118. https://doi.org/10.1063/5.0250559

[104] Kathuria, A., Lopez-Lengowski, K., Jagtap, S. S., McPhie, D., Perlis, R. H., Cohen, B. M., Karmacharya, R. Transcriptomic landscape and functional characterization of induced pluripotent stem cell–derived cerebral organoids in schizophrenia. *JAMA Psychiatry*, **2020**, 77(7): 745. https://doi.org/10.1001/jamapsychiatry.2020.0196

[105] Notaras, M., Lodhi, A., Dündar, F., Collier, P., Sayles, N. M., Tilgner, H., Greening, D., Colak, D. Schizophrenia is defined by cell-specific neuropathology and multiple neurodevelopmental mechanisms in patient-derived cerebral organoids. *Molecular Psychiatry*, **2022**, 27(3): 1416–1434. https://doi.org/10.1038/s41380-021-01316-6

[106] Walsh, R. M., Crabtree, G. W., Kalpana, K., Jubierre, L., Koo, S. Y., Ciceri, G., Gogos, J. A., Kruglikov, I., Studer, L. Forebrain assembloids support the development of fast-spiking human PVALB+ cortical interneurons and uncover schizophrenia-associated defects. *Neuron*, **2025**, 113(19): 3185–3203.e7. https://doi.org/10.1016/j.neuron.2025.06.017

[107] Zhang, W. D., Zhang, M., Xu, Z. H., Yan, H. Y., Wang, H. M., Jiang, J. M., Wan, J., Tang, B. S., Liu, C. Y., Chen, C. et al. Human forebrain organoid-based multi-omics analyses of PCCB as a schizophrenia associated gene linked to GABAergic pathways. *Nature Communications*, **2023**, 14: 5176. https://doi.org/10.1038/s41467-023-40861-2

[108] Nascimento, J. M., Saia-Cereda, V. M., Zuccoli, G. S., Reis-de-Oliveira, G., Carregari, V. C., Smith, B. J., Rehen, S. K., Martins-de-Souza, D. Proteomic signatures of schizophrenia-sourced iPSC-derived neural cells and brain organoids are similar to patients' postmortem brains. *Cell & Bioscience*, **2022**, 12: 189. https://doi.org/10.1186/s13578-022-00928-x

[109] Viswanath, B., Rao, N. P., Narayanaswamy, J. C., Sivakumar, P. T., Kandasamy, A., Kesavan, M., Mehta, U. M., Venkatasubramanian, G., John, J. P., Mukherjee, O. et al. Discovery biology of neuropsychiatric syndromes (DBNS): A center for integrating clinical medicine and basic science. *BMC Psychiatry*, **2018**, 18: 106. https://doi.org/10.1186/s12888-018-1674-2

[110] Qian, X. Y., Nguyen, H. N., Song, M. M., Hadiono, C., Ogden, S. C., Hammack, C., Yao, B., Hamersky, G. R., Jacob, F., Zhong, C. et al. Brain-region-specific organoids using mini-bioreactors for modeling ZIKV exposure. *Cell*, **2016**, 165(5): 1238–1254. https://doi.org/10.1016/j.cell.2016.04.032

[111] Wang, L., Nakamura, Y., Li, J. H., Sievert, D., Liu, Y., Nguyen, T., Jetti, P. S., Thai, E., Zhou, R. Y., Weng, J. M. et al. A phenotypic brain organoid atlas and biobank for neurodevelopmental disorders. *Cell Stem Cell*, **2025**, 32(12): 1923–1940.e7. https://doi.org/10.1016/j.stem.2025.10.006

[112] Yoon, J. G., Jang, H., Lee, S., Jang, S. S., Park, S., Cho, J., Kim, M., Han, J. Y., Yun, H., Kim, M. J. et al. Contribution of rare coding variants to microcephaly in individuals with neurodevelopmental disorders. *Genome Medicine*, **2025**, 17: 86. https://doi.org/10.1186/s13073-025-01513-w

[113] Maletic, M., Bizzotto, S., Ribierre, T., Guerdoud, K., Raoux, C., Doladilhe, M., Dalle, C. et al. Mosaic human cortical organoids model mTOR-related focal cortical dysplasia through *DEPDC5* deletion. *Brain*, **2026**: awag086. https://doi.org/10.1093/brain/awag086

[114] Kim, M. S., Kim, D. H., Kang, H. K., Kook, M. G., Choi, S. W., Kang, K. S. Modeling of hypoxic brain injury through 3D human neural organoids. *Cells*, **2021**, 10(2): 234. https://doi.org/10.3390/cells10020234

[115] Li, Y., Sun, X. Y., Zeng, P. M., Luo, Z. G. Neural responses to hypoxic injury in a vascularized cerebral organoid model. *Neuroscience Bulletin*, **2025**, 41(10): 1779–1791. https://doi.org/10.1007/s12264-025-01396-2

[116] Gaston-Breton, R., Disdier, C., Hagberg, H., Mabondzo, A. Hypoxia-ischemia and sexual dimorphism: Modeling mitochondrial dysfunction using brain organoids. *Cell & Bioscience*, **2025**, 15: 67. https://doi.org/10.1186/s13578-025-01402-0

[117] Paulsen, B., Velasco, S., Kedaigle, A. J., Pigoni, M., Quadrato, G., Deo, A. J., Adiconis, X., Uzquiano, A., Sartore, R., Yang, S. M. et al. Autism genes converge on asynchronous development of shared neuron classes. *Nature*, **2022**, 602(7896): 268–273. https://doi.org/10.1038/s41586-021-04358-6

[118] Fleck, J. S., Jansen, S. M. J., Wollny, D., Zenk, F., Seimiya, M., Jain, A., Okamoto, R., Santel, M., He, Z. S., Camp, J. G. et al. Inferring and perturbing cell fate regulomes in human brain organoids. *Nature*, **2023**, 621(7978): 365–372. https://doi.org/10.1038/s41586-022-05279-8

[119] Tamada, K., Takumi, T. Neurodevelopmental impact of CNV models in ASD: Recent advances and future directions. *Current Opinion in Neurobiology*, **2025**, 92: 103001. https://doi.org/10.1016/j.conb.2025.103001

[120] Fontenla, M. C. R., Pacoret, P. C., Alonso, S. D., Peñas, J. G., Parellada, M., Arango, C., Carracedo, A. Large-scale sequencing study of *de novo* regulatory Tandem Repeats (TRs) identifies new ASD (Autism Spectrum Disorders) candidate genes integrating

gene expression mapping, brain scRNA-seq and organoid models. *Research Square*, **2026**.

[121] Trudler, D., Ghatak, S., Bula, M., Parker, J., Talantova, M., Luevanos, M., Labra, S., Grabauskas, T., Noveral, S. M., Teranaka, M. et al. Dysregulation of miRNA expression and excitation in MEF2C autism patient hiPSC-neurons and cerebral organoids. *Molecular Psychiatry*, **2025**, 30(4): 1479–1496. https://doi.org/10.1038/s41380-024-02761-9

[122] Samarasinghe, R. A., Miranda, O. A., Buth, J. E., Mitchell, S., Ferando, I., Watanabe, M., Allison, T. F., Kurdian, A., Fotion, N. N., Gandal, M. J. et al. Identification of neural oscillations and epileptiform changes in human brain organoids. *Nature Neuroscience*, **2021**, 24(10): 1488–1500. https://doi.org/10.1038/s41593-021-00906-5

[123] Steinberg, D. J., Aqeilan, R. I. WWOX-related neurodevelopmental disorders: Models and future perspectives. *Cells*, **2021**, 10(11): 3082. https://doi.org/10.3390/cells10113082

[124] Yang, Y. L., Cai, Y., Wang, S. Y., Wu, X. L., Shao, Z. C., Wang, X., Ding, J. Human cortical organoids with a novel SCN2A variant exhibit hyperexcitability and differential responses to anti-seizure compounds. *Neuroscience Bulletin*, **2025**, 41(11): 2010–2024. https://doi.org/10.1007/s12264-025-01429-w

[125] Salmanzadeh, H., Halliwell, R. F. Human stem cell-derived neural organoids for the discovery of antiseizure agents. *Receptors*, **2025**, 4(3): 12. https://doi.org/10.3390/receptors4030012

[126] Zayat, V., Kuczynska, Z., Liput, M., Metin, E., Rzonca-Niewczas, S., Smyk, M., Mazurczak, T., Goszczanska-Ciuchta, A., Leszczynski, P., Hoffman-Zacharska, D. et al. The generation of human iPSC lines from three individuals with dravet syndrome and characterization of neural differentiation markers in iPSC-derived ventral forebrain organoid model. *Cells*, **2023**, 12(2): 339. https://doi.org/10.3390/cells12020339

[127] Zhu, W. D., Wu, Y., Li, X., Yang, H. J., He, F. C., Ma, J., Wei, J. Y., Leng, L. A stroke organoids-multiomics platform to study injury mechanism and drug response. *Bioactive Materials*, **2025**, 44: 68–81. https://doi.org/10.1016/j.bioactmat.2024.09.038

[128] Xiao, D. L., Sun, Y., Yang, G. Y., Yan, W. X., Jiang, M. L., Qin, Z. L., Wang, Z. J., Gu, Y. W., Zhou, J. T., Tan, J. et al. Nonexpansive biodegradable matrix promotes blood vessel organoid development for neurovascular repair and functional recovery in ischaemic stroke. *Nature Biomedical Engineering*, **2025**. https://doi.org/10.1038/s41551-025-01550-1

[129] Shiravi, S., Yufa, A., Papavasileiou, P., Lotz, S., Murphy, D., Bertucci, T., Temple, S., Finan, J. D. A model of traumatic brain injury oligomerizes tau in cortical organoids and induces clinically relevant pathologies that synergize with*MAPT*Mutation. *Journal of Neurotrauma*, **2025**, 42(21–22): 2004–2018. https://doi.org/10.1177/08977151251374286

[130] Lai, J. D., Berlind, J. E., Fricklas, G., Lie, C., Urenda, J. P., Lam, K., Sta Maria, N., Jacobs, R., Yu, V., Zhao, Z. et al. KCNJ2 inhibition mitigates mechanical injury in a human brain organoid model of traumatic brain injury. *Cell Stem Cell*, **2024**, 31(4): 519–536.e8. https://doi.org/10.1016/j.stem.2024.03.004

[131] Smirnova, L., Johnson, E. C. DROIDp – Drug Research Organoid-Integrated Development Platform: a Combinatorial NAM for Assessment of in vitro Learning and Memory in Drug and Chemical Testing. Research Portfolio Online Reporting Tools (RePORT), **2026**.

[132] Ramani, A., Müller, L., Ostermann, P. N., Gabriel, E., Abida‐Islam, P., Müller‐Schiffmann, A., Mariappan, A., Goureau, O., Gruell, H., Walker, A., et al. SARS-CoV-2 targets neurons of 3D human brain organoids. *EMBO J*, **2020**. 39(20): e106230. https://doi.org/10.15252/embj.2020106230

[133] Aguado, J., Amarilla, A. A., Taherian Fard, A., Albornoz, E. A., Tyshkovskiy, A., Schwabenland, M., Chaggar, H. K., Modhiran, N., Gómez-Inclán, C., Javed, I. et al. Senolytic therapy alleviates physiological human brain aging and COVID-19 neuropathology. *Nature Aging*, **2023**, 3(12): 1561–1575. https://doi.org/10.1038/s43587-023-00519-6

[134] Chen, C. W., Zhang, L. D., Dutta, R., Niroula, A., Miller, P. G., Gibson, C. J., Bick, A. G., Reyes, J. M., Lee, Y. T., Tovy, A. et al. SRCAP mutations drive clonal hematopoiesis through epigenetic and DNA repair dysregulation. *Cell Stem Cell*, **2024**, 31(2): 275–277. https://doi.org/10.1016/j.stem.2024.01.001

[135] Yoon, S. J., Elahi, L. S., Paşca, A. M., Marton, R. M., Gordon, A., Revah, O., Miura, Y., Walczak, E. M., Holdgate, G. M., Fan, H. C. et al. Reliability of human cortical organoid generation. *Nature Methods*, **2019**, 16(1): 75–78. https://doi.org/10.1038/s41592-018-0255-0

[136] Paşca, S. P., Arlotta, P., Bateup, H. S., Camp, J. G., Cappello, S., Gage, F. H., Knoblich, J. A., Kriegstein, A. R., Lancaster, M. A., Ming, G. L. et al. A nomenclature consensus for nervous system organoids and assembloids. *Nature*, **2022**, 609(7929): 907–910. https://doi.org/10.1038/s41586-022-05219-6

[137] Chiaradia, I., Lancaster, M. A. Brain organoids for the study of human neurobiology at the interface of *in vitro* and *in vivo*. *Nature Neuroscience*, **2020**, 23(12): 1496–1508. https://doi.org/10.1038/s41593-020-00730-3

[138] Durens, M., Nestor, J., Williams, M., Herold, K., Niescier, R. F., Lunden, J. W., Phillips, A. W., Lin, Y. C., Dykxhoorn, D. M., Nestor, M. W. High-throughput screening of human induced pluripotent stem cell-derived brain organoids. *Journal of Neuroscience Methods*, **2020**, 335: 108627. https://doi.org/10.1016/j.jneumeth.2020.108627

[139] Sirenko, O., Parham, F., Dea, S., Sodhi, N., Biesmans, S., Mora-Castilla, S., Ryan, K., Behl, M., Chandy, G., Crittenden, C. et al. Functional and mechanistic neurotoxicity profiling using human iPSC-derived neural 3D cultures. *Toxicological Sciences*, **2019**, 167(1): 58–76. https://doi.org/10.1093/toxsci/kfy218

[140] Cho, A. N., Jin, Y., An, Y., Kim, J., Choi, Y. S., Lee, J. S., Kim, J., Choi, W. Y., Koo, D. J., Yu, W. et al. Microfluidic device with brain extracellular matrix promotes structural and functional maturation of human brain organoids. *Nature Communications*, **2021**, 12: 4730. https://doi.org/10.1038/s41467-021-24775-5

[141] Albanese, A., Swaney, J. M., Yun, D. H., Evans, N. B., Antonucci, J. M., Velasco, S., Sohn, C. H., Arlotta, P., Gehrke, L., Chung, K. Multiscale 3D phenotyping of human cerebral organoids. *Scientific Reports*, **2020**, 10: 21487. https://doi.org/10.1038/s41598-020-78130-7

[142] Alam El Din, D. M., Shin, J., Lysinger, A., Roos, M. J., Johnson, E. C., Shafer, T. J., Hartung, T., Smirnova, L. Organoid intelligence for developmental neurotoxicity testing. *Frontiers in Cellular Neuroscience*, **2024**, 18: 1480845. https://doi.org/10.3389/fncel.2024.1480845

[143] Cryan, J. F., O'Riordan, K. J., Cowan, C. S. M., Sandhu, K. V., Bastiaanssen, T. F. S., Boehme, M., Codagnone, M. G., Cussotto, S., Christine, F., and Anna, V. G. The microbiota-gut-brain axis. *Physiological Reviews*, **2013**, 99(4): 1877–2013. https://doi.org/10.1152/physrev.00018.2018

[144] Workman, M. J., Mahe, M. M., Trisno, S., Poling, H. M., Watson, C. L., Sundaram, N., Chang, C. F., Schiesser, J., Aubert, P., Stanley, E. G. et al. Engineered human pluripotent-stem-cell-derived intestinal tissues with a functional enteric nervous system. *Nature Medicine*, **2017**, 23(1): 49–59. https://doi.org/10.1038/nm.4233

[145] Birey, F., Andersen, J., Makinson, C. D., Islam, S., Wei, W., Huber, N., Fan, H. C., Metzler, K. R. C., Panagiotakos, G., Thom, N. et al. Assembly of functionally integrated human forebrain spheroids. *Nature*, **2017**, 545(7652): 54–59. https://doi.org/10.1038/nature22330

[146] Andersen, J., Revah, O., Miura, Y., Thom, N., Amin, N. D., Kelley, K. W., Singh, M., Chen, X. Y., Thete, M. V., Walczak, E. M., et al. Generation of functional human 3D cortico-motor assembloids. *Cell*, **2020**, 183(7): 1913–1929.E26. https://doi.org/10.1016/j.cell.2020.11.017

[147] Skardal, A., Murphy, S. V., Devarasetty, M., Mead, I., Kang, H. W., Seol, Y. J., Shrike Zhang, Y., Shin, S. R., Zhao, L., Aleman, J. et al. Multi-tissue interactions in an integrated three-tissue organ-on-a-chip platform. *Scientific Reports*, **2017**, 7: 8837. https://doi.org/10.1038/s41598-017-08879-x

[148] Edington, C. D., Chen, W. L. K., Geishecker, E., Kassis, T., Soenksen, L. R., Bhushan, B. M., Freake, D., Kirschner, J., Maass, C., Tsamandouras, N. et al. Interconnected microphysiological systems for quantitative biology and pharmacology studies. *Scientific Reports*, **2018**, 8: 4530. https://doi.org/10.1038/s41598-018-22749-0

[149] Sharf, T., van der Molen, T., Glasauer, S. M. K., Guzman, E., Buccino, A. P., Luna, G., Cheng, Z. W., Audouard, M., Ranasinghe, K. G., Kudo, K. et al. Functional neuronal circuitry and oscillatory dynamics in human brain organoids. *Nature Communications*, **2022**, 13: 4403. https://doi.org/10.1038/s41467-022-32115-4

[150] Trujillo, C. A., Gao, R., Negraes, P. D., Gu, J., Buchanan, J., Preissl, S., Wang, A., Wu, W., Haddad, G. G., Chaim, I. A., et al. Complex oscillatory waves emerging from cortical organoids model early human brain network development. *Cell Stem Cell*, **2019**, 25(4): 558–569.e7. https://doi.org/10.1016/j.stem.2019.08.002

[151] Trevino, A. E., Sinnott-Armstrong, N., Andersen, J., Yoon, S. J., Huber, N., Pritchard, J. K., Chang, H. Y., Greenleaf, W. J., Pașca, S. P. Chromatin accessibility dynamics in a model of human forebrain development. *Science*, **2020**, 367(6476): eaay1645. https://doi.org/10.1126/science.aay1645

[152] Madhavan, M., Nevin, Z. S., Shick, H. E., Garrison, E., Clarkson-Paredes, C., Karl, M., Clayton, B. L. L., Factor, D. C., Allan, K. C., Barbar, L. et al. Induction of myelinating oligodendrocytes in human cortical spheroids. *Nature Methods*, **2018**, 15(9): 700–706. https://doi.org/10.1038/s41592-018-0081-4

[153] Marton, R. M., Miura, Y., Sloan, S. A., Li, Q. Y., Revah, O., Levy, R. J., Huguenard, J. R., Pașca, S. P. Differentiation and maturation of oligodendrocytes in human three-dimensional neural cultures. *Nature Neuroscience*, **2019**, 22(3): 484–491. https://doi.org/10.1038/s41593-018-0316-9

[154] Ormel, P. R., Vieira de Sá, R., van Bodegraven, E. J., Karst, H., Harschnitz, O., Sneeboer, M. A. M., Johansen, L. E., van Dijk, R. E., Scheefhals, N., Berdenis van Berlekom, A. et al. Microglia innately develop within cerebral organoids. *Nature Communications*, **2018**, 9: 4167. https://doi.org/10.1038/s41467-018-06684-2

[155] Schafer, S. T., Mansour, A. A., Schlachetzki, J. C. M., Pena, M., Ghassemzadeh, S., Mitchell, L., Mar, A., Quang, D., Stumpf, S., Ortiz, I. S., et al. An *in vivo* neuroimmune organoid model to study human microglia phenotypes. An *in vivo* neuroimmune organoid model to study human microglia phenotypes. *Cell*, **2023**, 186(10): 2111–2126.E20. https://doi.org/10.1016/j.cell.2023.04.022

[156] Park, D. S., Kozaki, T., Tiwari, S. K., Moreira, M., Khalilnezhad, A., Torta, F., Olivié, N., Thiam, C. H., Liani, O., Silvin, A. et al. iPS-cell-derived microglia promote brain organoid maturation *via* cholesterol transfer. *Nature*, **2023**, 623(7986): 397–405. https://doi.org/10.1038/s41586-023-06713-1

[157] Pham, M. T., Pollock, K. M., Rose, M. D., Cary, W. A., Stewart, H. R., Zhou, P., Nolta, J. A., Waldau, B. Generation of human vascularized brain organoids. *NeuroReport*, **2018**, 29(7): 588–593. https://doi.org/10.1097/wnr.0000000000001014

[158] Sloan, S. A., Darmanis, S., Huber, N., Khan, T. A., Birey, F., Caneda, C., Reimer, R., Quake, S. R., Barres, B. A., Pașca, S. P. Human astrocyte maturation captured in 3D cerebral cortical spheroids derived from pluripotent stem cells. *Neuron*, **2017**, 95(4): 779–790.e6. https://doi.org/10.1016/j.neuron.2017.07.035

[159] Choe, M. S., Kim, J. S., Yeo, H. C., Bae, C. M., Han, H. J., Baek, K., Chang, W., Lim, K. S., Yun, S. P., Shin, I. S. et al. A simple metastatic brain cancer model using human embryonic stem cell-derived cerebral organoids. *The FASEB Journal*, **2020**, 34(12): 16464–16475. https://doi.org/10.1096/fj.202000372r

[160] Pine, A. R., Cirigliano, S. M., Nicholson, J. G., Hu, Y., Linkous, A., Miyaguchi, K., Edwards, L., Singhania, R., Schwartz, T. H., Ramakrishna, R. et al. Tumor microenvironment is critical for the maintenance of cellular states found in primary glioblastomas. *Cancer Discovery*, **2020**, 10(7): 964–979. https://doi.org/10.1158/2159-8290.cd-20-0057

[161] López-Otín, C., Blasco, M. A., Partridge, L., Serrano, M., Kroemer, G. Hallmarks of aging: An expanding universe. *Cell*, **2023**, 186(2): 243–278. https://doi.org/10.1016/j.cell.2022.11.001

[162] Bussian, T. J., Aziz, A., Meyer, C. F., Swenson, B. L., van Deursen, J. M., Baker, D. J. Clearance of senescent glial cells prevents tau-dependent pathology and cognitive decline. *Nature*, **2018**, 562(7728): 578–582. https://doi.org/10.1038/s41586-018-0543-y

[163] Smirnova, L. Organoid intelligence (OI)–The ultimate functionality of a brain microphysiological system. *Altex*, **2023**: 191–203. https://doi.org/10.14573/altex.2303261

[164] Kagan, B. J., Kitchen, A. C., Tran, N. T., Habibollahi, F., Khajehnejad, M., Parker, B. J., Bhat, A., Rollo, B., Razi, A., Friston, K. J. *In vitro* neurons learn and exhibit sentience when embodied in a simulated game-world. *Neuron*, **2022**, 110(23): 3952–3969.e8. https://doi.org/10.1016/j.neuron.2022.09.001

[165] Cai, H. W., Ao, Z., Tian, C. H., Wu, Z. H., Liu, H. C., Tchieu, J., Gu, M. X., Mackie, K., Guo, F. Brain organoid computing for artificial intelligence. bioRxiv, **2023**: 2023.02.28.530502. https://doi.org/10.1101/2023.02.28.530502

[166] Musk, E., Neuralink. An integrated brain-machine interface platform with thousands of channels. *Journal of Medical Internet Research*, **2019**, 21(10): e16194. https://doi.org/10.2196/16194

[167] Willett, F. R., Kunz, E. M., Fan, C. F., Avansino, D. T., Wilson, G. H., Choi, E. Y., Kamdar, F., Glasser, M. F., Hochberg, L. R., Druckmann, S., et al. A high-performance speech neuroprosthesis. *Nature*, **2023**, 620(7976): 1031–1036. https://doi.org/10.1038/s41586-023-06377-x

[168] Cen, S., Gebregziabher, M., Moazami, S., Azevedo, C. J., Pelletier, D. Toward precision medicine using a "digital twin" approach: Modeling the onset of disease-specific brain atrophy in individuals with multiple sclerosis. *Scientific Reports*, **2023**, 13: 16279. https://doi.org/10.1038/s41598-023-43618-5

[169] Venkatesh, K. P., Raza, M. M., Kvedar, J. C. Health digital twins as tools for precision medicine: Considerations for computation, implementation, and regulation. *npj Digital Medicine*, **2022**, 5: 150. https://doi.org/10.1038/s41746-022-00694-7

[170] Schuman, C. D., Kulkarni, S. R., Parsa, M., Mitchell, J. P., Date, P., Kay, B. Opportunities for neuromorphic computing algorithms and applications. *Nature Computational Science*, **2022**, 2(1): 10–19. https://doi.org/10.1038/s43588-021-00184-y

[171] Jeziorski, J., Brandt, R., Evans, J. H., Campana, W., Kalichman, M., Thompson, E., Goldstein, L., Koch, C., Muotri, A. R. Brain organoids, consciousness, ethics and moral status. *Seminars in Cell & Developmental Biology*, **2023**, 144: 97–102. https://doi.org/10.1016/j.semcdb.2022.03.020

[172] Sidhaye, J., Knoblich, J. A. Brain organoids: An ensemble of bioassays to investigate human neurodevelopment and disease. *Cell Death & Differentiation*, **2021**, 28(1): 52–67. https://doi.org/10.1038/s41418-020-0566-4

[173] Lovell-Badge, R., Anthony, E., Barker, R. A., Bubela, T., Brivanlou, A. H., Carpenter, M., Charo, R. A., Clark, A., Clayton, E., Cong, Y. L. et al. ISSCR guidelines for stem cell research and clinical translation: The 2021 update. *Stem Cell Reports*, **2021**, 16(6): 1398–1408. https://doi.org/10.1016/j.stemcr.2021.05.012

[174] Tononi, G., Boly, M., Massimini, M., Koch, C. Integrated information theory: From consciousness to its physical substrate. *Nature Reviews Neuroscience*, **2016**, 17(7): 450–461. https://doi.org/10.1038/nrn.2016.44

[175] Koplin, J. J., Savulescu, J. Moral limits of brain organoid research. *Journal of Law, Medicine & Ethics*, **2019**, 47(4): 760–767. https://doi.org/10.1177/1073110519897789

[176] Savulescu, J., Sawai, T. Animus: Human-embodied animals. *Journal of Medical Ethics*, **2024**, 50(11): 725–728. https://doi.org/10.1136/jme-2022-108817

[177] Cleeremans, A., Mudrik, L., Seth, A. K. Consciousness science: Where are we, where are we going, and what if we get there? *Frontiers in Science*, **2025**, 3: 1546279. https://doi.org/10.3389/fsci.2025.1546279

[178] Albantakis, L., Barbosa, L., Findlay, G., Grasso, M., Haun, A. M., Marshall, W., Mayner, W. G. P., Zaeemzadeh, A., Boly, M., Juel, B. E. et al. Integrated information theory (IIT) 4.*0: Formulating the properties of phenomenal existence in physical terms. PLoS Computational Biology*, **2023**, 19(10): e1011465. https://doi.org/10.1371/journal.pcbi.1011465

[179] Casali, A. G., Gosseries, O., Rosanova, M., Boly, M., Sarasso, S., Casali, K. R., Casarotto, S., Bruno, M. A., Laureys, S., Tononi, G. et al. A theoretically based index of consciousness independent of sensory processing and behavior. *Science Translational Medicine*, **2013**, 5(198): e3006294. https://doi.org/10.1126/scitranslmed.3006294

[180] National Science and Technology Ethics Committee. Guiding Ethical Principles for Human Organoid Research. **2025**. Available at https://www.most.gov.cn/kjbgz/202504/P020250729547205340128.pdf

[181] Zhou, L. B., Lei, Y. T., Han, X. X. Charting bioethical frontiers: China's human organoid guidelines in a global context. *Military Medical Research*, **2025**, 12: 64. https://doi.org/10.1186/s40779-025-00651-x

[182] Andrews, M. G., Kriegstein, A. R. Challenges of organoid research. *Annual Review of Neuroscience*, **2022**, 45: 23–39. https://doi.org/10.1146/annurev-neuro-111020-090812